SECTION 5

SAMPLING METHODS

5.1 Introduction

5.1.1 Aquatic macroinvertebrates are good indicators of environmental water quality in fresh, estuarine, and marine waters. The analysis of faunal assemblages is an excellent way to detect water quality problems. Different kinds of stress will often produce different communities of benthic macroinvertebrates. The sampling equipment and methods discussed can be used to study and analyze macroinvertebrate communities for ambient or special studies, and the resulting data and information can be used to document both spatial and temporal changes in water quality. The sampling devices and methods of this section relate to qualitative, semi-quantitative, and quantitative sampling.

5.1.1.1 Qualitative and semi-quantitative sampling of macroinvertebrates are relatively easy. The current methodology discussed here is well developed, and the equipment needed for sampling is not elaborate. Many effective methods of data analysis, including pollution indices and diversity indices, have been developed for use with macroinvertebrates (also, see Section 7, Data Evaluation).

5.1.1.2 Quantitative sampling is more difficult. Random sampling and the patchy distribution of macroinvertebrates within the substrate often means larger numbers of samples are needed in order to be able to make reasonable estimates of community structure and population densities. However, this is not a problem confined only to macroinvertebrates, but to other aquatic animals as well. Also, see Section 4, Selection of Sampling Sites and Section 7, Data Evaluation.

5.1.2 The sampling methods employed should depend on the data quality objectives (DQOs) (see Section 2, Quality Assurance and Quality Control) of the study determined by interaction of the decision making authority and biomonitoring expertise of qualified aquatic biologists.

5.1.3 A list of equipment, supplies, and companies that can provide sampling gear for collecting benthic macroinvertebrates can be found in Appendix E.

5.2 Qualitative Sampling

5.2.1 The objective of qualitative studies is to make within or between site comparisons to determine the presence or absence of benthic macroinvertebrates having varying degrees of tolerance to pollution and to obtain information on the richness of taxa, at or near the species level (taxa present and relative abundance). Samples are obtained with the use of a wide variety of collecting methods and gear, many of which are not amenable to quantification on a unit-area or volume basis. Any collecting device (e.g., dip or hand nets, kick nets, screens, dredges, grab samplers, stream-net samplers, and artificial substrate samplers) can be used for qualitative collections of macroinvertebrates. The use of several methods of collection at each station can increase the total number of taxa collected. When conducting qualitative studies, an attempt is usually made to collect as many taxa as possible in the time available by exhaustive sampling in all available habitat types. No habitat should be overlooked at the site if the objective of the study is to obtain a representative collection of the macroinvertebrates.

5.2.2 Experience and skill are required in selecting suitable collecting techniques and recognizing and locating various types of habitats where qualitative samples can be collected.

5.2.3 When conducting comparative studies of the macrobenthos, a major drawback is the confounding effect of the differences in physical habitat among the different stations being studied. This problem is particularly inherent in qualitative studies when an attempt is made to systematically collect as many species as possible at the sampling stations or reaches of streams being compared. Unfortunately, differences in habitat unrelated to the effects of pollution may render such comparisons meaningless. To minimize this drawback, the investigator should carefully record, in the field, the habitats from which specimens are collected (a habitat assessment) and then base comparisons only on stations with like habitats in which the same amount of collecting effort has been expended. Appropriate sampling methods, such as the use of artificial substrates, should be utilized to eliminate the problem of comparing different physical habitats among stations being studied.

5.2.4 Advantages of qualitative sampling are the wide latitude in collecting methods, the types of habitats that can be sampled are relatively unrestricted, and the processing of the samples is usually less time consuming.

5.2.5 Limitations of qualitative sampling include collecting techniques that are subjective and depend on the skill and experience of the sample collector, sampling results of one investigator can be difficult to compare with those of another, and no information on standing crop or biomass can be generated from a qualitative study.

5.3 Semi-quantitative Sampling

5.3.1 Semi-quantitative sampling data can be generated based on methods that measure the collection of benthic macroinvertebrates by level of effort (e.g., time expended per habitat) or when quantitative sampling devices are used to collect samples in a non-random manner. Examples of some semi-quantitative methods include the 10 rock method (Lewis, personal communication), traveling kick method (Hornig and Pollard, 1978; Pollard, 1981), and Rapid Bioassessment Protocols II and III (Plafkin *et al*., 1989). See Section 7, Data Evaluation.

5.4 Quantitative Sampling

5.4.1 Quantitative methods essentially provide an estimation of the numbers or biomass (standing crop) of the various components of the macroinvertebrate community per unit area, volume, or sampling unit. The method also provides information on the species composition, richness of species, and distribution of individuals among the species. The high variability often associated with some macroinvertebrate populations makes them difficult to study quantitatively (Schwenneker and Hellenthal, 1984), but multi-metric assessment endpoints are used to avoid the difficulty of utilizing only population-based measurement endpoints. Section 7, Data Evaluation and Elliott (1971) should provide statistical principles for sampling and data analyses of benthic macroinvertebrates.

5.4.2 Quantitative estimates are obtained by using devices that sample a unit area or volume of the habitat. The major considerations are the size of the sampling units, the number of sampling units in each sample, and the location of sampling units in the sampling area. Grab samplers, stream-net samplers (e.g., Surber and related type samplers, Hess and related type samplers, and drift nets), and artificial substrate type samplers, are examples of devices that are used to collect samples quantitatively.

5.4.3 Sampling precision in the study of macroinvertebrate populations is affected by the substrate area encompassed by the sampling device and the patchiness in distribution of the organisms. The smaller the substrate surface area encompassed by a sampling device, the larger the number of sampling units required to obtain a desired level of precision (Elliott, 1971). Precision can be increased by collecting larger sampling units or by increasing the numbers of sampling units collected. A quantitative approach necessitates that a measure of the precision be obtained by replicate sampling. Replicate sampling in each habitat (habitat niche, microhabitat, or strata) selected for study is an absolute requirement.

5.4.3.1 For measurement of precision, three replicate random sampling units at each sampling station are an absolute minimum. Five replicates at each station would increase the statistical precision and accuracy. A series of single sampling units taken at discrete points along a transect do not represent replicate samples of benthic organisms unless it can be demonstrated that the physical characteristics of the habitat do not change along the transect.

5.4.4 The total number of samples depends on the degree of precision required, which will depend on the type of study and data quality objectives (DQOs). A reconnaissance or pilot study of the station may be necessary to help determine the sample size. Southwood (1966) gives a formula for determining the number of sampling units required for a specific level of precision.

5.4.5 The data from properly designed quantitative studies are amenable to the use of simple but powerful statistical tools that aid in maintaining the objectivity of the data evaluation process (see Section

7, Data Evaluation). The measures of precision and probability statements that can be attached to quantitative data reduce the possibilities of bias in the data evaluation process and make the results of different investigators more readily comparable. The advantages of quantitative methods are that they provide a measure of invertebrate diversity, biomass, and productivity, and their associated precision, thereby providing objective comparisons within, between, and among studies or intra- and interstudy comparisons.

5.4.6 No one sampling device is completely adequate to sample all types of habitat. When either qualitative, semi-quantitative, or quantitative devices are used, only selected portions of the environment are usually sampled. Also, because of the potential use of these data, experienced and skilled biologists are needed for sample collections.

5.5 Sampling Devices

5.5.1 Grab Samplers (Grabs)

5.5.2 Grabs are devices designed to penetrate the substrate by virtue of their own weight and leverage and have spring- or gravity-activated closing mechanisms. The jaws of grabs are forced shut by weights, lever arms, springs, or cables. All grabs are designed to take discrete "bites" or "scoops" of a defined area into the bottom sediment of a lake, stream, estuary, ocean, or similar habitats to sample the benthos. Scoops are grab samplers that scoop sediment with a rotating container. In shallow waters, some of these devices may be rigged on poles or rods and physically pushed into the substrate to a predetermined depth.

5.5.2.1 The number and kinds of macroinvertebrates collected by a particular grab may be affected by the habitat sampled, substrate type sampled, depth of penetration, angle of closure, completeness of closure of the jaws and loss of sample material during retrieval, creation of a "shock" wave and consequent "washout" of organisms at the surface of the substrate, and the effect of the high-flow velocities often encountered in rivers and wave action in large lakes and oceans on the stability of the sampler.

5.5.2 Selecting Grab Sampling Devices

5.5.2.1 Table 3 summarizes criteria for selecting grabs.

TABLE 3. SUMMARY CRITERIA FOR GRAB SAMPLERS

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1. Ponar Grab (Standard)

   A. Habitats and Substrates Sampled: Freshwater lakes, rivers, estuaries, and reservoirs with hard and soft sediments such as clay, hard pan, sand, gravel and muck; somewhat less efficient in softer sediments.

B. Effectiveness of the Device: Not entirely adequate for deep burrowing organisms in soft sediments; very efficient for hard sediments; collects both qualitative and quantitative samples.

C. Advantages: Better penetration than other grabs; side plates and screens reduce washout, shock waves and substrate disturbance; best quantitative grab sampler for freshwater use.

D. Limitations: A very heavy grab that requires use of a boat with winch and cable; stones, pebbles and other debris can hold jaws open causing loss of sample.

2. Petite Ponar Grab

A. Habitats and Substrates Sampled: Freshwater lakes, rivers and reservoirs and estuaries with moderately hard sediments such as sand, silt and mud; will not penetrate clay; somewhat less efficient in soft sediments and coarse gravel.

B. Effectiveness of the Device: Not entirely adequate for deep burrowing organisms in soft sediments; not useful in clay.

C. Advantages: Good penetration for such a small grab; side plates and screens reduce washout, shock waves and substrate disturbance; can be operated by hand without boat or winch.

D. Limitations: Jaws can be blocked by stones, sticks and other debris causing loss of part of the sample; not efficient in swiftly flowing water of over one meter per second velocity.

Selected Literature: APHA, 1989; ASTM, 1990; Brinkhurst, 1967, 1974; Elliott _et al_., 1978, 1980, 1981b; Flannagan, 1970; Howmiller, 1971; Hudson, 1970; Lewis _et al_., 1982; Powers and Robertson, 1967; USEPA, 1973.

3. Ekman Grab (Standard, Tall, Large, and Extra-large)

A. Habitats and Substrates Sampled: Freshwater rivers, lakes and reservoirs where there is little current; soft sediments such as muck and silt.

B. Effectiveness of the Device: Efficient only in soft sediments but weights can be added for deeper penetration in fine sand; collects both qualitative and quantitative samples.

C. Advantages: Easy to operate by hand without winch, can be pushed into substrate in shallow water; hinged doors at top reduce washout, shock waves and disturbance of the substrate; comes in a range of sizes.

D. Limitations: Light weight so that jaw will not penetrate hard substrates; jaws often do not close completely due to blocking of jaws or failure of closing mechanism; inefficient in deep water or where there is even moderate current.

Wildco box corer resembles a heavy duty Ekman grab that has been designed to penetrate harder substrates with the addition of a frame and weights. The device can be used to collect infauna of lakes and estuaries. The box corer may also be used to sample finely divided muck, clays, mud, ooze, submerged marl, or fine peaty bottoms. The sampler weighs about 14 kg, but a maximum of 49 kg (12 removable weights) may be used. The sample area is 150 x 150 x 225 mm; a removable acrylic liner is included.

Selected Literature: APHA, 1989; ASTM, 1990; Beattie, 1979; Burton and Flannagan, 1973; Ekman, 1911, 1947; Flannagan, 1970; Howmiller, 1971; Hudson, 1970; Lanz, 1931; Lewis et al., 1982; Lind, 1974; Milbrink and Wiederholm, 1973; Paterson and Fernando, 1971; Rowe and Clifford, 1973; Rawson, 1947; Schwoerbel, 1970; Welch, 1948; USEPA. 1973.

4. Petersen Grab (Standard and Baby)

A. Habitats and Substrates Sampled: Freshwater lakes, reservoirs and rivers and estuaries with sand, gravel, clay and hard pan substrates.

B. Effectiveness of the Device: Less effective in most substrates than the Ponar, Baby Petersen effective in moderately soft sediments.

C. Advantages: Can give quantitative samples if used properly; range of sizes available.

D. Limitations: Standard grab is heavy and requires boat with winch; can cause washout if dropped rapidly to the bottom; shallow bite by jaws so that deeper burrowing organisms are not sampled; jaws are easily blocked by debris causing loss of sample; hard to use in adverse weather; of questionable value as a quantitative sampler.

Selected Literature: APHA, 1989; ASTM, 1990; Barnes, 1959; Birkett, 1958; Brinkhurst, 1974; Davis, 1925; Edmondson and Winberg, 1971; Elliott and Tullett, 1978; Holme and McIntyre, 1971; Hudson, 1970; Howmiller, 1971; Jensen, 1981; Lewis et al., 1982; Petersen, 1918; Petersen and Tensen, 1911.

5. Smith-McIntyre Grab

A. Habitats and Substrates Sampled: Marine and estuaries; adaptable to large rivers, lakes and reservoirs with sand, gravel, clay and similar substrates.

B. Effectiveness of the Device: Limited penetration; has been widely used for sampling in marine and estuarine habitats.

C. Advantages: Provides reasonably quantitative samples; trigger plates help penetrate the substrate.

D. Limitations: Very heavy, needs boat and power winch; spring loaded jaws could be hazardous; inefficient for collecting deep burrowing organisms; jaws can be blocked by debris.

Selected Literature: APHA, 1989; ASTM, 1990; Carey and Heyamoto, 1972; Carey and Paul, 1968; Elliott and Tullett, 1978; Holme, 1964; Hopkins, 1964; Hunter and Simpson, 1976; McIntyre, 1971; Smith and McIntyre, 1954; Tyler and Shackley, 1978; Wigley, 1967; Word, 1976, 1977; Word et al., 1976.

6. Van Veen Grab

A. Habitats and Substrates Sampled: Marine and estuaries with sand, gravel, mud, clay and similar substrates; could be adapted to freshwater.

B. Effectiveness of the Device: Penetrates to a depth of 5 to 7 cm.

C. Advantages: Jaws close better than the Petersen Grab; samples most types of sediments; comes in a range of sizes.

D. Limitations: A very heavy grab that requires a large boat and power winch; jaws may become blocked by debris such as rocks and sticks; not useful for deep burrowing organisms.

Selected Literature: APHA, 1989; ASTM, 1990; Barnes, 1959; Beukema, 1974; Birkett, 1958; Elliott and Drake, 1981b; Elliott and Tullett, 1978; Holme, 1971; Lassig, 1965; Longhurst, 1959; McIntyre, 1956; Nichols and Ellison, 1966; Schwoerbel, 1970; Ursin, 1954; Wigley, 1967; Word, 1976a, 1976b; Word et al., 1976.

7. Orange-Peel Grab

A. Habitats and Substrates Sampled: Marine waters and deep lakes with sandy substrates containing cobble, rubble and coarse gravel.

B. Effectiveness of the Device: For qualitative use only; sampling area not constant.

C. Advantages: Comes in a range of sizes; works well in deep water; closes relatively well to prevent loss of sample; good for reconnaissance.

D. Limitations: Very heavy so that large boat with power winch and cable lines is required; does not sample constant area and depth.

Selected Literature: APHA, 1989; ASTM, 1990; Briba and Reys, 1966; Elliott and Tullett, 1978; Hartman, 1955; Hopkins, 1964; Merna, 1962; Packard, 1918; Reish, 1959; Thorson, 1957, Word, 1976, 1977.

8. Shipek Grab

A. Habitats and Substrates Sampled: Estuaries and large deep lakes with sand, gravel, mud and clay substrates.

B. Effectiveness of the Device: A relatively good quantitative sampler.

C. Advantages: Good for collecting a small sample in deep water.

D. Limitations: A heavy grab that requires the use of a boat with a power winch; must be lowed vertically so is not effective in moving water; inefficient for collecting deep burrowing organisms; samples small area.

Selected Literature: APHA, 1989; ASTM, 1990; Barnes, 1959; Elliott and Tullett, 1978; Flannagan, 1970; Holme, 1964, 1971; Holme and McIntyre, 1971.

5.5.3 Precautions

5.5.3.1 Always inspect grabs for mechanical defects prior to use.

5.5.3.2 Exercise caution at all times when handling grabs.

5.5.4 Significance and Use of Grabs

5.5.4.1 Qualitative and quantitative samples of macroinvertebrates inhabiting sediments or substrates are may be taken by grabs. Grab samplers, if used correctly, are devices that sample a unit area of the habitat. In view of the advantages and limitations regarding the penetration of the sediment by many grabs and their closing mechanisms, it is not possible to recommend any single instrument as suitable for general use. However, the Petersen grab is considered the least effective bottom grab sampler and, therefore, has limited application. The type and size of the grab sampler or device selected for use will depend on such factors as the size of boat, hoisting gear available, the type of substrate or sediment to be sampled, depth of water, current velocity, and whether sampling is in sheltered areas or in open waters of large rivers, reservoirs, lakes, or oceans. The choice of grab will depend largely on what is available, what is suitable for the sampling area, and what can be used with the least difficulty.

5.6 Commonly Used Grabs

5.6.1 The ponar grab sampler (Fig. 5A,B) is most commonly used for sampling macroinvertebrates from sediments in lakes, rivers, reservoirs, estuaries, and oceans with coarse and hard substrates, such as coarse sand, gravel, and similar substrates, rather than soft sediments, such as mud, fine sand, or sludge. The sampler can be used in moderate currents and deep waters.

5.6.1.1 The Ponar grab sampler has paired jaws that must penetrate beneath the surface of the substrate without disturbing the water surface boundary layer, close when positioned properly on the bottom, and retain discrete samples of sediment while it is brought to the surface for processing. The device has side plates and a screen on the top of the sample compartment to prevent loss of the sample during closure. With one set of weights, this heavy steel sampler can weigh 20 Kg. Word _et al_. (1976a) reports that the large amount of surface disturbance associated with Ponar grabs can be greatly reduced by simply installing hinges rather than fixed screen tops, which will reduce the pressure wave associated with the sampler's descent into the sediment. The standard Ponar takes a sample area of 523 cm^2. A small version, the petite Ponar grab, takes a sample area of 232 cm^2 and can be used in habitats where there may be an unusual abundance of macroinvertebrates, thus eliminating the need to subsample.

5.6.1.2 When not in use, a safety pin lock attached to the lever bar prevents closing of the sampler until the pin is removed.

5.6.1.3 The weight of the standard Ponar grab makes it necessary to use a winch and cable or portable crane for retrieving the sample, and ideally the samples should be taken from a stationary boat or platform. The smaller version, petite Ponar grab, is designed for hand-line operation, but it may be used with a winch and cable.

5.6.2 The Ekman grab sampler (Fig. 5C) is used to obtain samples of macroinvertebrates from soft sediments, such as very fine sand, mud, silt, and sludge, in lakes, reservoirs, estuaries, and similar habitats where there is little current. This grab is inefficient in deep waters, under adverse weather conditions, and in waters of moderate to strong currents or wave action. The Wildco box corer (Fig. 5D) is like a heavy duty Ekman with a frame and weights and is used to collect macroinvertebrates in lakes and estuaries. Because of its weight a winch is necessary for retrieving the sample from a stationary boat or platform.

5.6.2.1 The Ekman grab sampler is a box-shaped device with two scoop-like jaws that must penetrate the intended substrate without disturbing

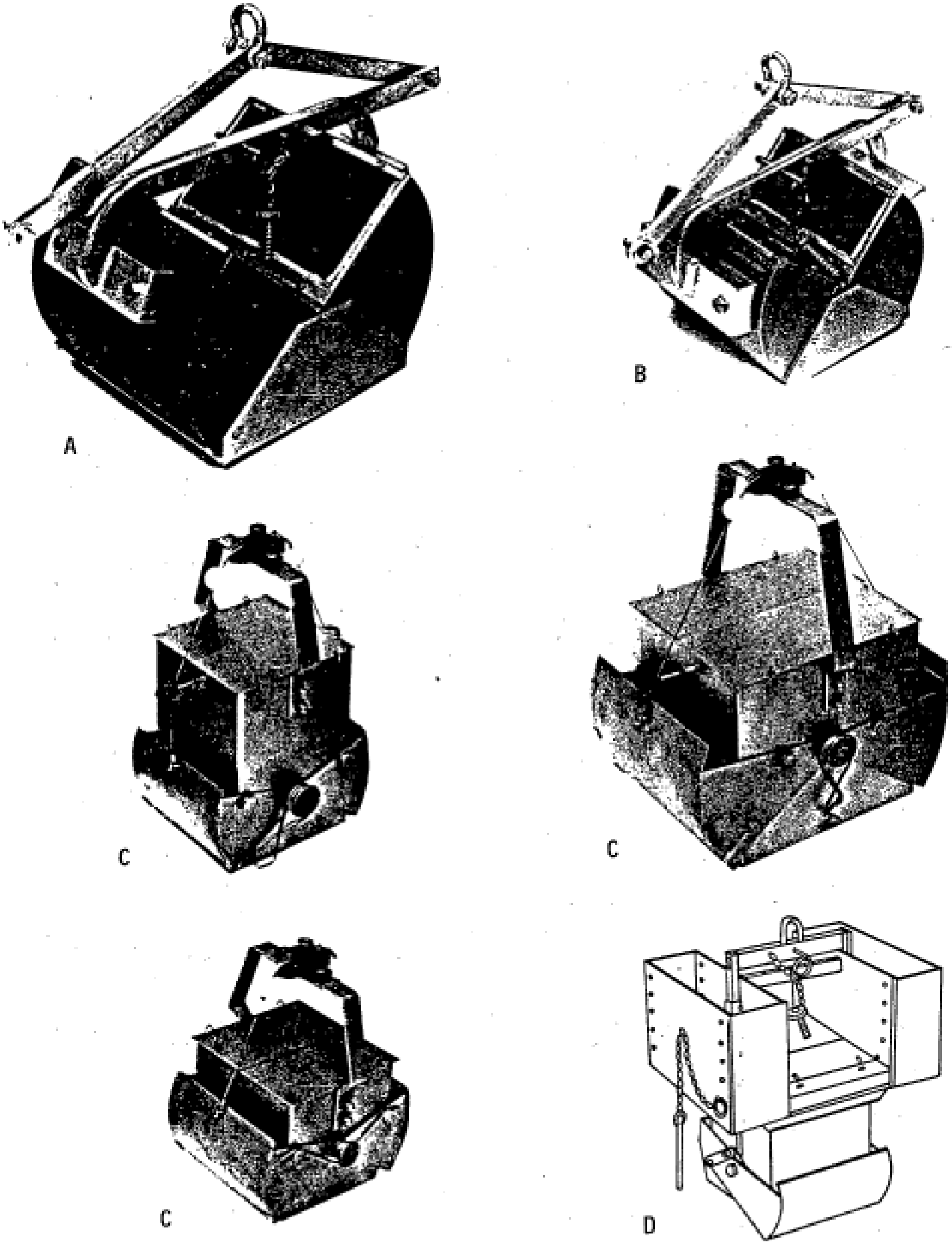

Figure 5. Grab Samplers. (A) Standard Ponar; (B) Petite Ponar; (C) Large, tall, and standard Ekman grabs; (D) Wildco box corer

the water surface boundary layer, close when positioned properly on the bottom, and retain a discrete sample of sediment while it is brought to the surface for processing. Hinged doors on the top of the grab prevents washout during sample lowering and retrieval. The grab is made of 12 to 20 gauge brass or stainless steel and weighs approximately 3.2 Kg. The box-like part holding the sample has spring-operated jaws on the bottom that must be manually set. The sampler is available in several sizes; however, in very soft substrates only a tall model should be used, either a 23 cm or a 30.5 cm model. Ekman is not used with a winch very often but can be operated from a boat with a winch and cable.

5.6.2.2 Exercise caution at all times once the grab is loaded or cocked because a safety lock is not part of the standard design.

5.6.3 The Petersen grab sampler (Fig. 6A,B) is designed to obtain samples of macroinvertebrates from sediments in lakes, reservoirs, and similar habitats and is adaptable to rivers, estuaries, and oceans. This grab sampler has limited application, and is not recommended for quantitative benthic work and must be used with due consideration of its defects when quantitative estimates are attempted. It is useful for sampling sand, gravel, marl, and clay in moderate currents and deep waters, the sampler cannot be used under adverse weather conditions. This sampler is available in a range of sizes that will sample an area from 0.06 to 0.099 m^2. A consensus of aquatic biologists consider the use of this device the least preferable grab sampler and would use it only in limited applications.

5.6.3.1 The Petersen grab sampler has paired jaws that must penetrate the intended substrate without disturbing the water surface boundary layer, close when positioned properly on the bottom, and retain the sample of sediment while it is brought to the surface for processing. This heavy steel device can weigh 13.7 Kg, but may weigh as much as 31.8 Kg when auxiliary weights are bolted to its side. The extra weights are to make the grab stable in swift current and to give additional cutting force in firm bottom sediments. It has been suggested that users of this device modify it by the addition of end plates and by cutting large strips out at the top of each side and adding hinged 30 mesh screen as in the Ponar grab. It is necessary to use a winch and cable to lower and raise the sampler.

5.6.3.2 Newer versions of the Petersen grab sampler may have a screened window at the top of each jaw to allow water to escape while the grab is descending and closing. While some modifications may close or function better, the sampling characteristics remain the same. Most of the modified versions are intended for use in estuarine and marine waters.

5.6.3.3 Ideally a stationary boat or platform should be used when taking samples. The modified Petersen devices are designed to be quite heavy and require heavy gear and a large vessel for efficient operation. A small version can be hauled aboard by hand and held with one hand for washing procedures.

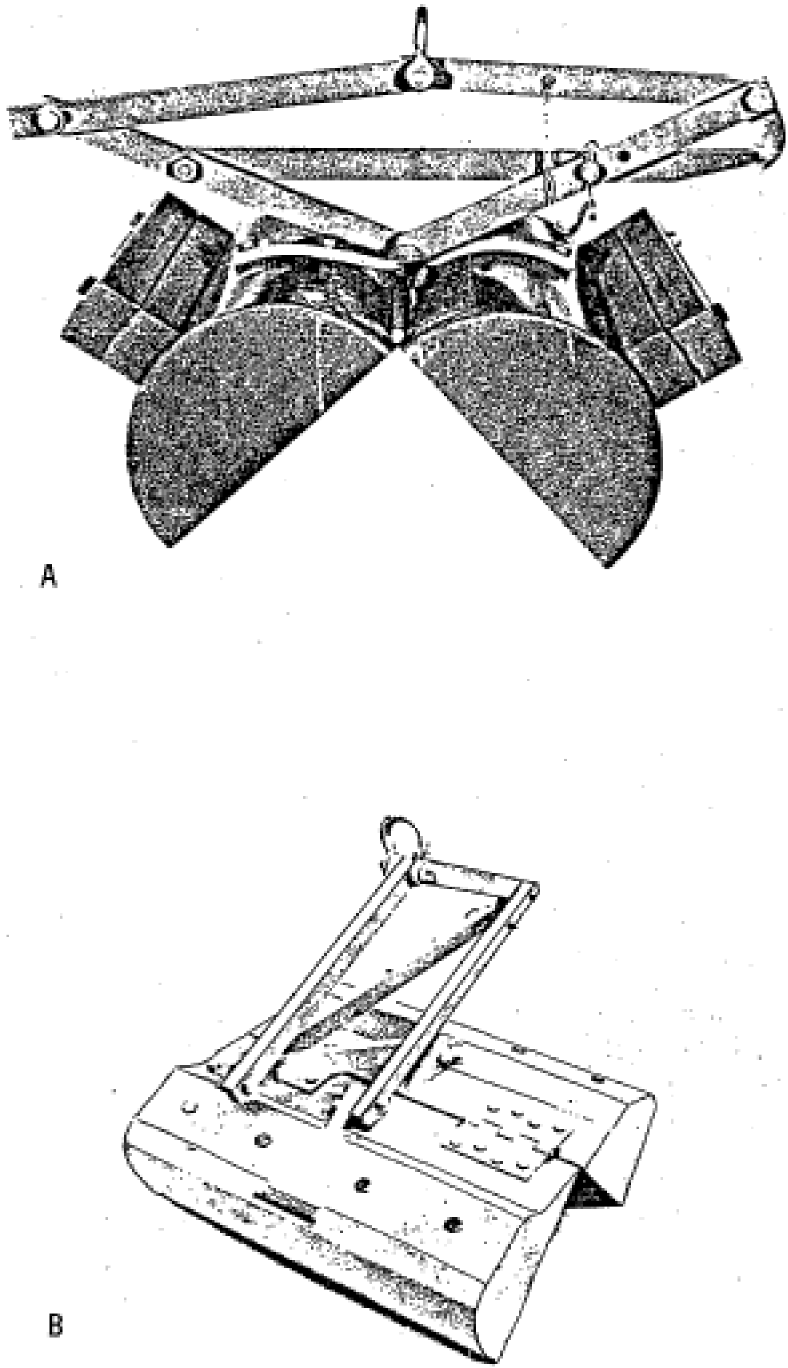

Figure 6. Grab Samplers: (A) Original Petersen; (B) Modified Petersen

5.6.4 The Smith-McIntyre grab sampler (Fig. 7A) is designed to obtain samples of macroinvertebrates from sediments in rough weather and deep water in lakes, rivers, estuaries, and oceans. This device samples a surface area of 0.1 m^2 and is useful for sampling macroinvertebrates from sand, gravel, mud, clay, and similar substrates.

5.6.4.1 The Smith-McIntyre grab sampler has paired jaws that are forced to penetrate into the intended substrate by two "loaded" strong coiled springs, must close when positioned properly on the bottom, and retain discrete samples of sediment while it is brought to the surface for processing. The device is heavy and can weigh 45.4 Kg or more. The chief advantages of the sampler are its stability and easier control in deep and rough waters. The spring-loaded jaws of the Smith-McIntyre grab must be considered a hazard and caution should be exercised when using the device. Due to the weight and size, this device must be used from a vessel with boom and lifting capabilities.

5.6.4.2 The Smith-McIntyre grab sampler is fitted with gauze panels or free swinging panels on the top to reduce the shock wave during descent.

5.6.4.3 Larger Smith-McIntyre grabs can be constructed depending on the type of bottom to be sampled and additional weights can be fitted to the frame of the grab sampler for additional penetration into the sediment.

5.6.5 The Van Veen grab sampler (Fig. 7B) is used to obtain samples of macroinvertebrates from sediments in estuaries and other marine habitats, and is adaptable to freshwater areas. It can also be used for qualitative sampling. This device is useful for sampling sand, gravel, mud, clay and similar substrates and is available in two sizes, 0.1 m^2 and 0.2 m^2. Larger and double versions of this grab are available, and their use is dependent upon the type of bottom to be sampled, and the type of vessel available to deploy this sampler.

5.6.5.1 The Van Veen grab sampler has paired jaws that must penetrate the intended substrate without disturbing the water surface boundary layer of the substrate, close by pincher-like action of two long arms when positioned properly on the bottom, and retain discrete samples of sediment while it is brought to the surface for processing. The long arms give added leverage for penetrating hard sediments. The advantage of using the twin Van Veen is that with a single lowering, two separate bottom sediment sampling units can be collected from the same station.

5.6.5.2 The Van Veen is basically an improved version of the Petersen grab in that long arms have been attached to the jaws to stabilize the grab on the bottom in the open sea just prior to or during closure of the device. Additional weights can be applied to the jaws to effect greater penetration in sediments.

5.6.6 The Orange-Peel grab sampler (Fig. 7C) is used primarily in marine waters and deep lakes where it has advantages over other grabs when sandy substrates are sampled, but it cannot be used under adverse weather conditions. This grab should not be used in critical quantitative work that is to be compared with results of other areas and is recommended as a reconnaissance sampler only. The sampler is available in a range of sizes but the 1600 cm^3 is generally used, although larger sizes are available.

5.6.6.1 The Orange-Peel grab sampler has four curved jaws that close to encircle a hemisphere of sediment. It must penetrate the intended substrate without disturbing the water surface boundary layer, close when positioned properly on the bottom, and retain discrete samples of sediment while it is brought to the surface for processing. The top of the sampler is enclosed by a canvas bag, serving as a portion of the sample compartment. When taking samples, a stationary boat or platform should be used.

5.6.6.2 A recent modification of the Orange-Peel, described by Reish (1959) has a new trigger mechanism and more efficient closing jaws, and the volume of sample to surface-area sampled relationship has been worked out.

5.6.6.3 The surface area sampled by this device varies with penetration depth or volume sampled. The device penetrates to a maximum depth of 18 cm, but depth of penetration will vary.

5.6.7 The Shipek (scoop) grab sampler (Fig. 7D) is designed to obtain samples of macroinvertebrates from sediments in marine waters and large inland bodies of water. This device is useful for sampling macroinvertebrates from sand, gravel, mud, clay, and similar substrates. It is designed to take a sediment sample with a surface area of 20 cm^2 to approximately 10 cm deep at the center.

5.6.7.1 The Shipek (scoop) grab sampler consists of a semi-cylindrical scoop that must be positioned properly on the bottom to take a scoop and retain discrete samples of sediment through 180°. Holmes and McIntyre (1971) report that this device is usually used by geologists to collect small samples rather than by biologists. However, it can be used in marine waters and large inland lakes, reservoirs, and rivers. Unlike many other types of samplers, closure of the device is made at the side, rather than at the bottom. This sampler cannot be used under adverse wind and wave conditions. The sampler requires a vessel with a winch and cable.

5.6.8 General Operating Procedures

5.6.8.1 Most grabs are heavy sampling devices that should be operated using a hand or powered winch and cable from a boat. In large bodies of water ships are used for this operation.

5.6.8.2 Grabs must be lowered slowly because free-fall may airplane the device, causing the device to land improperly or causing a pressure wave and blowout of the surface layer of sediment when the grab reaches the

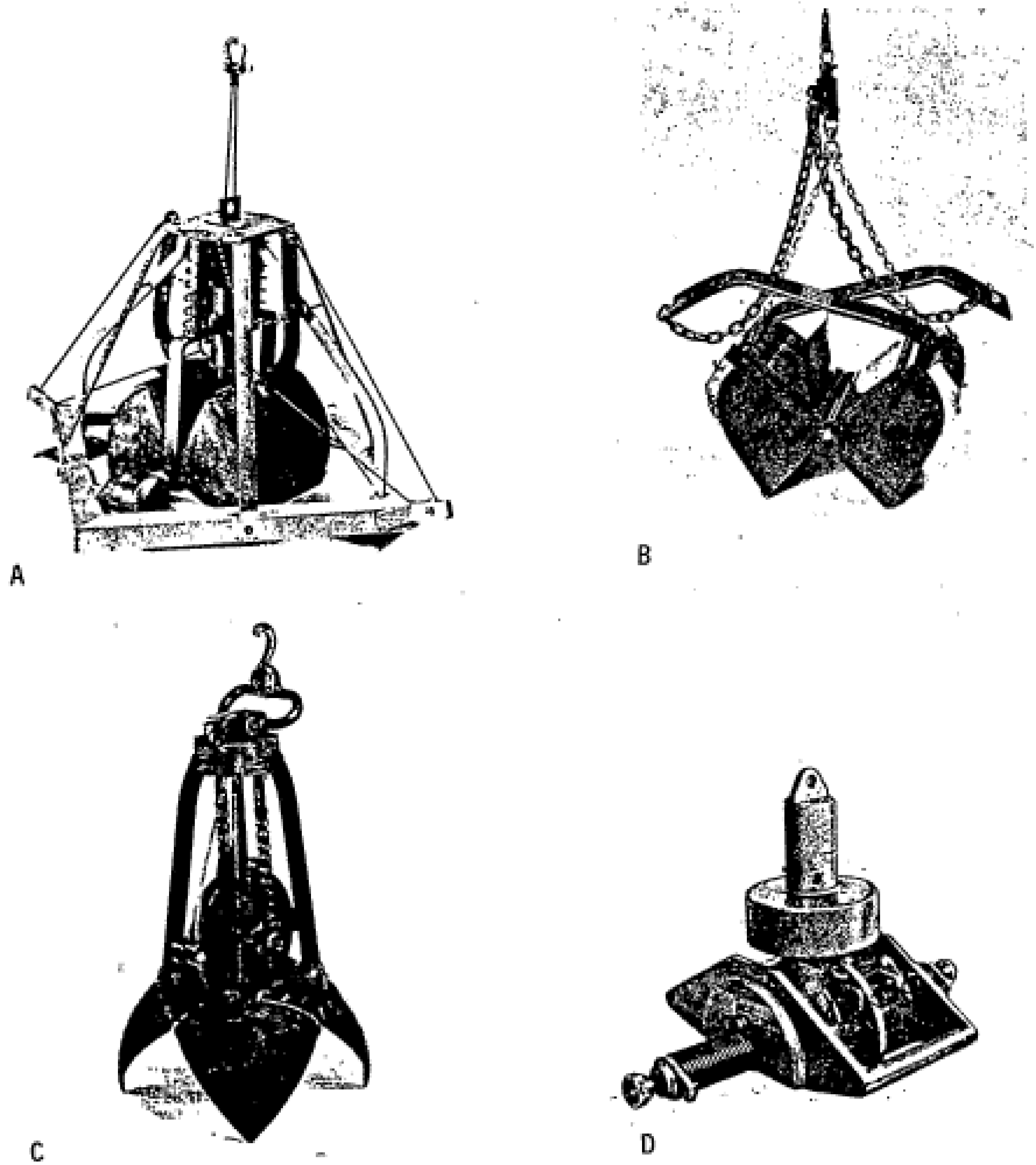

Figure 7. Grab Samplers: (A) Smith-McIntyre; (B) Van Veen; (C) Orange-Peel; (D) Shipek

bottom. In order for the device to operate effectively, it must bite vertically.

5.6.8.3 When most grabs reach the bottom, their weight will cause them to penetrate the substrate, and the slack-off on the cable allows the locking lever to release, therefore permitting the movement that allows the horizontal locking bar to drop out of the locking notch and allow the jaws to close as the device is raised. Other grabs are closed by spring action or some other mechanical device after penetrating the substrate.

5.6.8.4 In the Ekman grab the jaws are cocked by raising them upward into the cocked position using the attached cable and securing the cable to the catch pin located at the top of the sampler. Once on the bottom, indicated by a slack line, a messenger is sent down the line tripping the catch mechanism, causing the spring loaded jaws to close the bottom of the sampler and contain the sediment.

5.6.8.5 The Smith-McIntyre grab is "loaded" by compressing the large coil springs mounted on the instrument with the loading bar. As soon as the spring is loaded, the safety pin is inserted to prevent the accidental triggering of the bottom plates. Once the device is overboard, just prior to lowering to the bottom, the safety pins are removed. When the trigger plates contact the bottom, pressure on these plates releases the two coiled springs that drive the buckets (jaws) into the sediment. Closure of the sampler is made at the side, rather than at the bottom. After closure the sample is given optimum protection from washout during the return trip to the surface by the cylindrical configuration of the sampler. Once on deck, the sampler is placed on a stand; the sample buckets can be disengaged from the rest of the device by releasing two retaining latches at each end of the upper semicylinder, and the sample is dumped into a large basin or washtub and prepared for processing. After the sample has been removed, the springs may then be loaded and the safety pins installed.

5.6.8.6 The chains from the jaws of the Van Veen are attached to the counter balance mechanism, as are the slackened wires from the long arms. Tension is carefully applied to the trigger mechanisms as the sampler is winched off its platform, and once the tension is firmly changed from the jaws, the grab is relatively stable in the cocked position. Care should be exercised in lowering the Van Veen through the surface of the water as occasionally contact will produce slack in the chain that will trip the counter balance mechanism. The grab is lowered slowly to the bottom, and once it makes contact with the bottom, the grab is winched in initially closing the jaws containing the sediment. Retrieve the grab slowly to prevent washout.

5.6.8.7 The Shipek grab is composed of two concentric half cylinders, the inner semicylinder is rotated at high torque by two spirally wound external springs. Upon contact with the bottom, the two external springs are automatically released by the inertia of a self-contained weight upon a sear mechanism which trips the catch and the scoop rotates upward. At the end of its 180° travel, the sample bucket is stopped and held at the closed position by residual spring torque. After closure the sample is given optimum protection from washout. The scoop is disengaged from the upper semicylinder by releasing the two retaining latches at each end of the upper semicylinder.

5.6.8.8 Once on board, the sample is placed into either a suitable container or a sieving device directly for processing (see Section 6). Thoroughly wash or hose the grab with water, so that all sediment materials are included in the sample before a replicate sample is taken.

5.7 Stream-Net Samplers

5.7.1 Stream-net samplers are lotic collecting devices, fitted with a net of various mesh sizes that collect organisms from flowing water passing through the sampler.

5.7.2 Selecting Stream-Net Sampling Devices

5.7.2.1 Table 4 summarizes criteria for selecting stream-net sampling devices.

TABLE 4. SUMMARY CRITERIA FOR STREAM-NET SAMPLERS

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1. Surber Sampler

   A. Habitats and Substrates Sampled: Shallow, flowing streams, less than 32 cm in depth with good current; rubble substrate, mud, sand, gravel.

   B. Effectiveness of Device: Relatively quantitative when used by experienced biologist; performance depends on current and substrate.

   C. Advantages: Encloses area sampled; easily transported or constructed; samples a unit area.

   D. Limitations: Difficult to set in some substrate types, that is, large rubble; cannot be used efficiently in still, slow moving streams.

2. Portable Invertebrate Box Sampler, Hess Sampler, Hess Stream Bottom Sampler, and Stream-Bed Fauna Sampler

   A. Habitats and Substrates Sampled: Same as Surber.

   B. Effectiveness of Device: Same as Surber.

   C. Advantages: Same as above except completely enclosed with stable platform; can be used in weed beds.

TABLE 4. SUMMARY CRITERIA FOR STREAM-NET SAMPLERS (continued)

D. Limitations: Same as Surber.

Selected Literature: APHA, 1989; ASTM, 1990; Canton and Chadwick, 1984; Elliott and Tullett, 1978; Ellis and Rutter, 1973; Hess, 1941; Kroger, 1972; Lane, 1974; Merritt et al., 1984; Needham and Usinger, 1956; Pollard and Kinney, 1979; Rutter and Ellis, 1977; Rutter and Poe, 1978; Rutter and Ettinger, 1977; Resh, 1979; Resh et al., 1984; Schwenneker and Hellenthal, 1984; Surber, 1937, 1970; Usinger, 1963; Waters and Knapp, 1961; Welch, 1948; Winner et al., 1980.

3. Drift Nets

A. Habitats and Substrates Sampled: Flowing rivers and streams; all substrate types.

B. Effectiveness: Relatively quantitative and effective in collecting all taxa which drift in the water column; performance depends on current velocity and sampling period.

C. Advantages: Low sampling error; less time, money, effort; collects macroinvertebrates from all substrates, usually collects more taxa.

D. Limitations: Unknown where organisms come from; terrestrial species may make up a large part of sample in summer and periods of wind and rain; does not collect non-drifting organisms.

Selected Literature: Allan, 1984; Allan and Russek, 1985; APHA, 1989, ASTM, 1990; Bailey, 1964; Berner, 1951; Brittain and Eikeland, 1988; Chaston, 1969; Clifford, 1972a,b; Coutant, 1964; Cushing, 1963, 1964; Dimond, 1967; Edington, 1965; Elliott, 1965, 1967; 1969, 1970; 1971; Elliott and Minshall, 1968; Ferrington, 1984; Hales and Gaufin, 1969; Hemsen, 1956; Hildebrand, 1974; Holt and Waters, 1967; Hynes, 1970; Keefer and Maughan, 1985; Larimore, 1972, 1974; Larkin and McKone, 1985; Lehmkuhl and Anderson, 1972; McLay, 1970; Merritt et al., 1984; Minshall and Winger, 1968, Modde and Schulmbach, 1973, Muller, 1965, 1974, Mullican et al., 1967; Mundie, 1959, 1964; Pearson and Franklin, 1968; Pearson and Kramer, 1969, 1972; Pearson et al., 1968; Pfitzer, 1954; Radford and Hartland-Rowe, 1971; Reisen and Prins, 1972; Resh, 1979; Resh et al., 1984; Spence and Hynes, 1971; Tanaka, 1960; Tranter and Smith 1968; USEPA, 1973; Waters, 1961, 1962, 1964, 1965; 1966; 1968, 1969a,b, 1972; Wilson and Bright, 1973; Winner et al., 1980; Wojtalik and Waters, 1970.

5.7.3 The Surber, portable invertebrate box, Hess, Hess stream bottom, and stream-bed fauna samplers (Fig. 8A-E) were designed as quantitative samplers when carefully used by an experienced biologist; however, they are more often used to collect qualitative samples or semi-quantitative samples because of the large number of samples needed for an acceptable level of precision (Needham and Usinger, 1956). They outline a definite unit-area for collecting the macroinvertebrates within the area. They are designed to be placed by hand onto or in some cases into sand, gravel, or rubble substrate types (usually in riffle/run areas) in shallow streams, or shallow areas of rivers. The drift net sampler (Fig. 8F) is a qualitative and quantitative collecting device used to capture drifting organisms in flowing waters. It differs from the other net type samplers in that it collects from a unit volume of water rather than from a unit area of bottom.

5.7.4 Significance and Use of Stream-Net Samplers

5.7.4.1 The significance of using stream-net samplers is to collect macrobenthos inhabiting a wide range of habitat types from shallow flowing streams or shallow areas in rivers. The stream-net devices (Surber, portable invertebrate box, Hess, Hess stream bottom, and stream-bed fauna samplers) are unit area samplers used for collecting benthic organisms in certain types of substrates. They may be used to obtain estimates of the standing crop, for example, biomass, number of individuals and number of taxa of benthic macroinvertebrates per unit area of stream bottom. Efficiency of the sampler depends on the experience and ability of the user. Drift net samplers are designed to collect emigrating or dislodged benthic macroinvertebrates inhabiting all substrate types that either actively or passively enter the water column in flowing streams and rivers and is used to determine drift density and drift rate.

5.7.5 Description of Surber Type Samplers

5.7.5.1 The Surber sampler consists of two 30.5-cm frames, hinged together; one frame rests on the substrate, the other remains upright and holds the nylon net. The sampler is positioned with its net mouth open, facing upstream. When in use, the two frames are locked at right angles, one frame marking off the area of substrate to be sampled and the other frame supporting a net to strain out organisms washed into it from the sample area.

5.7.5.2 Modification of the Surber sampler to overcome some of the limitations of its use (for example, loss of organisms due to backwash) has resulted in the design and construction of a number of related sampling devices, such as the four-sided (enclosed) portable invertebrate box sampler, the cylindrical Hess sampler, the cylindrical Hess stream bottom sampler, and the cylindrical stream-bed fauna sampler. These devices sample 0.1 m^2.

5.7.5.3 Operation of the portable invertebrate box, Hess, Hess stream bottom, and stream-bed fauna samplers are similar to the Surber sampler.

5.7.5.4 The net used to collect macroinvertebrates can vary in mesh size, length, taper, and material, for example, canvas, taffeta, or

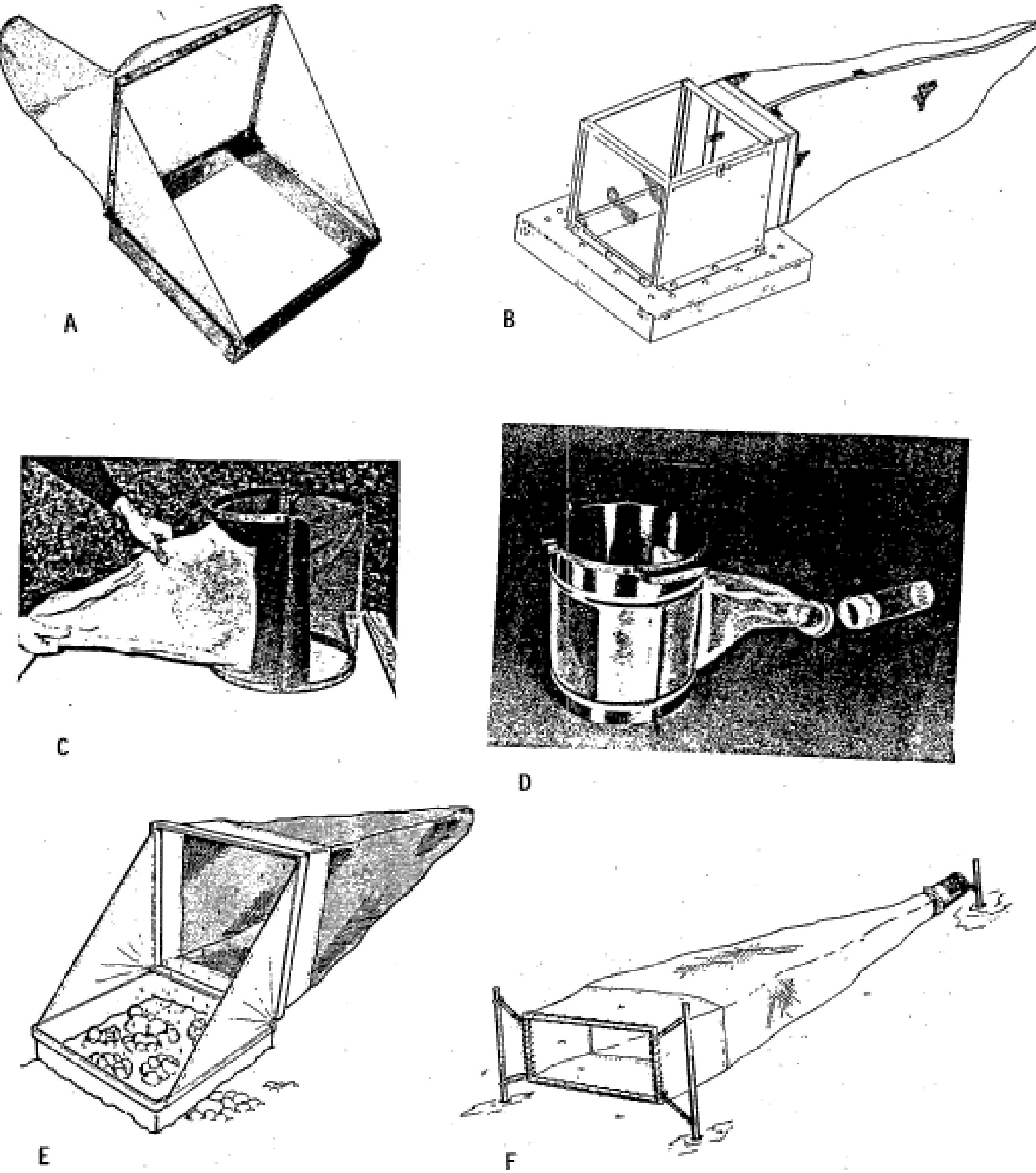

Figure 8. Stream-Net Samplers: (A) Surber sampler; (B) Portable invertebrate box sampler; (C) Hess sampler; (D) Hess stream bottom sampler; (E) Stream-bed fauna sampler (F) Drift net

nylon monofilament. It is usually made of nylon, and a variety of mesh sizes is available. The mesh size used will depend on the objectives of the study. A mesh size of 0.35 mm, for example, will retain most instars of aquatic insects.

5.7.5.6 While a smaller mesh size might increase the number of smaller invertebrates and young instars collected, it will clog more easily and exert more resistance to the current than a larger mesh, possibly resulting in a loss of organisms due to backwashing from the sample net.

5.7.5.7 The polyester foam base of the portable invertebrate box sampler conforms to a variety of substrates to prevent the loss of organisms from beneath the sampler. The Hess, Hess stream bottom, and stream-bed fauna samplers can be "turned" into most sediment types to a depth of several centimeters. The Surber sampler rests on the surface of most sediments.

5.7.5.8 When sampling is completed, the net of the portable invertebrate box sampler slides out for cleaning or exchange with a different net. Hess-type samplers may have a mason jar ring and an adapter with a fixed or removable cloth net bucket. Some of the stream-net samplers have fixed nets.

5.7.5.9 These samplers cannot be used as efficiently in still or deep water of more than 30.48 cm (1-ft) depth. If the water depth is greater than 30.48 cm (1-ft), benthic organisms may wash over the top of the net rather than into it.

5.7.5.10 While there can be large sampling errors associated with their use by an inexperienced operator, these samplers can provide data which are precise and comparable if they are used consistently by one experienced person in similar habitats.

5.7.5.11 If the water velocity is very great, resistance provided by the small mesh of the net or debris washed into it, or both, may result in a backwashing effect that washes benthic organisms out of the sample area of the Surber sampler or over the top of the other samplers.

5.7.6 General Operating Procedures

5.7.6.1 Position these samplers securely on the substrate, parallel to the flow of the water, with the net pointing downstream.

5.7.6.2 The samplers are brought down quickly to reduce the escape of rapidly moving organisms.

5.7.6.3 There should be no gaps under the edges of the frame that would allow for washing of water under the net and loss of benthic organisms. Eliminate gaps that may occur along the edge of the Surber sampler frame by careful shifting of rocks and gravel along the outside edge of the sampler. This is also true of the cylindrical-type samplers if they are on rubble substrate that makes turning into the bottom difficult. The portable invertebrate box sampler polyester foam pad can conform to a relief of 7.6 cm (3 in.).

5.7.6.4 Take care not to disturb the substrate upstream from the sampler, to avoid excessive drift into the sampler from outside the sample area.

5.7.6.5 Once the sampler is positioned on the stream bottom, it should be maintained in position during sampling so that the area delineated remains constant.

5.7.6.6 Hold the Surber sampler with one hand or brace with the knees from behind. The Hess, Hess stream bottom, and stream-bed fauna samplers, and the portable invertebrate box samplers can be held with one hand or braced with the knees from the sides. The portable invertebrate box sampler also can be sat upon for convenience while sampling; this provides the collector with a stable sampling platform that allows maximum manipulation of the substrate with little sampler movement.

5.7.6.7 Heavy gloves should be required when handling dangerous debris; for example, glass or other sharp objects present in the sediment.

5.7.6.8 Turn over and examine carefully all rocks and large stones and rub carefully in front of the net with the hands or a soft brush to dislodge the organisms and pupal cases, etc. clinging to them before discarding. Scrape attached algae, insect cases, etc., from the stones into the sample net.

5.7.6.9 Wash larger components of the substrate within the enclosure with stream water; water flowing through the sampler should carry dislodged organisms into the net.

5.7.6.10 Stir the remaining gravel and sand vigorously with the hands to a depth of 10 cm (4.0 in.) where applicable, depending upon the substrate, to dislodge bottom-dwelling organisms.

5.7.6.11 It may be necessary to hand pick some of the heavier mussels and snails that are not carried into the net by the current.

5.7.6.12 Remove the sample by inverting the net (or washing out sample bucket, if applicable) into the sample container (wide-mouthed jar) with 10% buffered formalin fixative or 70-80% ethanol.

5.7.6.13 Examine the net carefully for small organisms clinging to the mesh, and remove them (preferably with forceps to avoid damage) for inclusion in the sample.

5.7.6.14 Rinse the sampler net after each use.

5.8 Drift Nets

5.8.1 Significance and Use of Drift Nets

5.8.1.1 Macroinvertebrate drift is a normal feature of flowing waters (Brittain and Eikeland, 1988). Drift of organisms may be used to assess environmental stress or pollution in some situations. Stress, fluctuations in water level, changes in light intensity, and changes in temperature are the basic factors that influence the extent of macroinvertebrate drift.

5.8.1.2 One source of drifting macroinvertebrates is the immature insects in the final stages of metamorphosis that actively seek to reach the water surface where emergence to the adult stage occurs. Regular periodic downstream drift rate of immature insects and other macroinvertebrate fauna in slow-moving streams or rivers is markedly reduced in comparison to lotic habitats with rapidly flowing water.

5.8.2.3 Drift insects are about evenly distributed at all levels in a stream, but in large rivers drift is more abundant near the bottom in the shore-line zone.

5.8.2.4 It is generally found that there are pulses of drift organisms that move from top to bottom of the water column, at least during periods of low flow.

5.8.2.5 Drift collections can be used to determine drift density, rate, and periodicity of drift organisms, and interesting aspects of the organisms' life histories, for example, period of transformation.

5.8.2.6 Drift nets are useful for collecting macroinvertebrates that actively or passively enter the water column or that are dislodged from the substrate; naturally or by stress. They are particularly well-suited for synoptic surveys because they are light weight and easily transported.

5.8.2.7 The first step in interpreting drift data is to determine the respective contributions of constant, behavioral, and catastrophic drift to the samples being analyzed.

5.8.2.8 Only constant and behavioral drift are usually utilized in a synoptic survey, but catastrophic drift is extremely important in testing for recent discharges of toxic materials.

5.8.2.9 Bear in mind that the drift density may not be a function of the total bottom population density or of production; however, species composition of the drift is useful as an index of species composition of the benthos.

5.8.2.10 Density and composition of invertebrate drift are influenced by many factors that also must be considered when interpreting the data, including stage of life cycle, weather, time of day, light intensity, population density, temperature, turbidity, water level fluctuation, season, current velocity, growth rate, photoperiod, and proximity to tributary streams.

5.8.2.11 In an enriched stream there is usually a marked increase in total numbers and biomass of drifting organisms as the stream becomes more polluted. Intolerant forms decrease and pollution tolerant forms increase proportional to changing water quality.

5.8.2.12 Thousands of organisms, including larvae of stoneflies, mayflies, caddisflies, and midges and other Diptera, may be collected in a sampling period of only a few hours.

5.8.2.13 The drift net efficiently collects organisms originating from all types of substrates upstream and a wide spectrum of microhabitats in lotic (flowing) waters.

5.8.2.14 The device is restricted to flowing rivers or streams with a current velocity of more than 0.05 m/s.

5.8.3 Advantages of Using Drift Nets

5.8.3.1 A benthic sample shows only which taxa were existing in the particular area (usually some fraction of a square meter, etc.) that was sampled. The great variation among benthic samples, even in a limited area, illustrates the necessity of several samples and the influence of selecting the collecting stations. One drift sample might be adequate for collecting the majority of invertebrate taxa in a stream reach, whereas a large number of benthic samples would be needed to cover the variety of bottom habitats even in an uniform reach of the stream.

5.8.3.2 Quantitative benthic sampling is seldom extended to include stream banks, organic substrates (logs, etc.), and areas of dense vegetation. The drift net collects organisms from all these areas.

5.8.3.3 Drift net collections often require much less sorting work than a series of grab samples. Drift samples do not require the laborious, time-consuming job of washing out silts, clays, and other materials and of sorting and picking through much of the debris for the organisms in the samples.

5.8.3.4 Nets are light-weight and easy to set up in a stream and usually yield a light-weight sample free from most debris. Benthic sampling in flowing water often procures samples heavy with inorganic materials.

5.8.3.6 A drift net is inexpensive to construct, whereas bottom samplers are often costly and more than one kind may be required to adequately sample the multiple habitat types present in a stream or river.

5.8.4 Limitations of Use of Drift Nets

5.8.4.1 Certain aquatic organisms enter the drift only sporadically and might be missed even though common in the benthos.

5.8.4.2 The relative abundance of macroinvertebrates in a drift sample often differs significantly from their "relative" abundance on the stream bottom.

5.8.4.3 A slight current is necessary if a drift collection is to be taken (greater than 0.05 m/s).

5.8.4.4 Most species drift more abundantly at night, so that the best collections are usually taken in the dark. Time of sampling depends on the purpose of the study. Day samples are usually adequate for showing effects of pollution on the stream reach.

5.8.4.5 There is a waiting period while the drifting organisms accumulate in the net, but not as long as with using artificial substrates.

5.8.4.6 Tree leaves in the autumn, floating and anchor ice in the winter, and heavy debris (logs) during floods may interfere with drift net collecting and make processing difficult.

5.8.4.7 The abundance and composition of drift changes, daily, hourly, or seasonally and might prevent direct comparison of collections taken at different times. At times certain life stages of an organism might not be fairly represented in the drift. The same holds true for other types of sampling.

5.8.4.8 Drift collections give little precise habitat information for individual organisms, since the exact source of the individual is not known.

5.8.4.9 Collections of drift, with the organisms originating an indefinite distance above the collecting site, may not show local or temporary deleterious effects imposed on an aquatic community, whereas bottom samples might reveal the destruction or reduction of benthos in a small area. Studies have shown that most drift organisms originate from only several meters upstream from the nets (Elliott, 1967).

5.8.5 Description of Drift Nets

5.8.5.1 The typical drift net consists of a bag of nylon or nylon monofilament. The drift net generally preferred is the simple rectangular net which is light-weight, easy to install, and gives an adequate sample of the drifting macroinvertebrates. The U.S. Standard No. 30 (0.595-mm mesh openings) net is often used for collecting macroinvertebrates.

5.8.5.2 Drift nets vary in size, but the type recommended for use in water pollution surveys or other ecological assessments has an upstream opening of 15 by 30 cm, and the collection bag is 1.3 m long. A variety of mesh sizes is available, and mesh size should be selected based on the objectives of the study; the finer the mesh, the more organisms (instars) will be collected.

5.8.5.3 The frame typically consists of a 0.045-m^2 (15 by 30-cm) brass rod structure anchored into the stream bed by a pair of steel rods.

5.8.5.4 Drift nets are anchored in the stream by driving 1/2-in. steel rods into the stream bottom or mounting the rods in concrete slabs that are weighted down with stones. Use cable clamps to secure the nets to the rods.

5.8.5.5 The drift net frame can be fitted anteriorly with a mouth reducing rectangular plexiglass enclosure (Rutter and Ettinger 1977) to increase filtration efficiency and volume of water passing through the net.

5.8.5.6 Alternatives to the typical drift net include the waterwheel drift sampler (Pearson and Kramer, 1969) which might be useful in large rivers or streams with slow flow which can be reached by automobile.

5.8.5.7 An automatic drift sampler (Muller, 1965) can be constructed that eliminates the need of an attendant at the sampling site during collection of as many as eight consecutive samples.

5.8.5.8 A modified emergence-trap drift sampler (Mundie, 1964; Cushing, 1964) is useful in streams with extremely high drift, where water is very turbid, or where a long sampling period is desired without clogging.

5.8.5.9 The average volume of water passing through the net is determined by measuring the water velocity at the mouth of the drift net with a current meter at the beginning of the sampling period and at the end of the sampling period using the average, and recording the total time the drift net is set in the water column. Results are expressed as numbers per cm^3 of water passing through the net.

5.8.5.10 The efficiency of the net is determined by the simultaneous measurement of the water velocity passing by the set drift net.

5.8.6 General Operating Procedures

5.8.6.1 Because the performance and sampling efficiency of a drift net sampler varies with local stream conditions, seasonal changes, and water level, make a preliminary test before the start of regular drift sampling in order to determine the best sampling stations, best sampling interval, number of nets needed, mesh size, and best sampling depth.

5.8.6.2 For synoptic surveys, one net set above each of the major areas of population concentrations is usually adequate; but for definitive studies a minimum of two drift nets should be set at each station so that drift from above a pollution source, drift from the polluted reach, and drift from the zone of clean water downstream from the recovery zone can be compared.

5.8.6.3 Take into consideration the fact that the drift net will collect drifting organisms that may have entered the drift from an indefinite distance upstream or a tributary stream. Nets located 80 to 100 m below the effluent will generally sample the polluted reach efficiently. A drift net below a riffle collects more animals than one below a pool.

5.8.6.4 For definitive studies, install four nets at each station - two about 25 cm from the bottom and two about 10 cm below the surface in water not exceeding 3 m in depth.

5.8.6.5 If the objective of the study is to relate pupal exuviae to pollution, or to collect terrestrial organisms that may float on the surface, then extend one net slightly above the surface.

5.8.6.6 Ideally, collect 24-h drift samples; but this is usually not practicable unless one resorts to the use of a water-wheel, automatic drift sampler, or a modified drift sampler with a restricted opening to solve the clogging problem or by changing the nets at regular intervals.

5.8.6.7 Although the sampling interval will vary with time of day, current velocity, density of drift organisms, and floating debris, collect 1-3 hours daytime drift samples when either a 24-h or overnight sampling period is not prudent.

5.8.6.8 Drift nets have also been used from small boats in large rivers (Rutter and Ettinger, 1977).

5.8.6.9 Because the size of the catch varies as the flow of water through the net varies, it is necessary to measure the current velocity at the entrance of each net at the beginning and end of each sampling period so that the catch can be converted into number of organisms per volume of water flowing through the net.

5.8.6.10 At the end of the specified sampling period, remove the net from the water by loosening the cable clamps and raising the net over the top of the steel rods, taking care not to disturb the bottom upstream of the net.

5.8.6.11 Concentrate the material in the net in one corner by swishing up and down in the water and then wash into a bucket half-filled with water. Then sieve and handle the sample in the regular manner.

5.8.6.12 Subdividing the sample substantially reduces analysis time with large samples (Waters, 1969a and USEPA, 1973).

5.8.6.13 Reporting data as numbers of individuals per net is meaningless because no two drift net samples are collected under exactly the same conditions of current velocity, stream discharge, and sampling interval. Conversion equations and other statistical aspects of drift sampling are given by Elliott (1970). An equation for converting the data to number per 100 m^3 of water flow is:

$$X = 100a/bdc$$

where:
X = number of organisms per 100 m^3,
a = number of organisms in the net (density)
b = number of minutes of the sampling interval,
c = current velocity, m/min, and
d = area of the net opening in m^2.

5.9 Artificial Substrate Samplers

5.9.1 Artificial substrate samplers are devices made of natural or artificial materials of various composition and configuration that are placed in water for a predetermined period of exposure and depth for the colonization of indigenous macroinvertebrate communities. They are used in obtaining qualitative and quantitative samples of macroinvertebrates in rivers, streams, lakes, and reservoirs.

5.9.2 Artificial substrate sampling can effectively augment bottom substrate sampling because many of the physical variables encountered in bottom sampling are minimized (e.g., variable depth and light penetration, temperature differences, and substrate types).

5.9.3 Samples usually contain negligible amounts of extraneous material, permitting quick laboratory processing.

5.9.4 Selecting Artificial Substrate Samplers

5.9.4.1 Table 5 summarizes criteria for selecting artificial substrate samplers.

TABLE 5. SUMMARY CRITERIA FOR ARTIFICIAL SUBSTRATE SAMPLERS

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1. Multiplate (Modified Hester-Dendy) Sampler

   A. Habitats and Substrates Sampled: All types of habitats in rivers, streams, lakes and reservoirs; not efficient in wetlands; uses hardboard or porcelain substrate.

   B. Effectiveness of the Device: Colonization depends on type of substrate; selective for certain types of organisms; three replicates considered adequate.

   C. Advantages: Excellent for water quality monitoring; uniform substrate type; high level of precision; samples contain negligible amount of debris; provides habitats of known area for a known time at a known depth.

TABLE 5. SUMMARY CRITERIA FOR ARTIFICIAL SUBSTRATE SAMPLERS (Continued)

D. Limitations: Requires trip for installation and trip for collection; subject to vandalism; biased for aquatic insects; need to use caution in reuse of plates that may have been contaminated with toxicants, oil, etc.; may require additional weight for stability; up to eight weeks wait for results.

Selected Literature: APHA, 1989; Beck et al., 1973; Beckett and Miller, 1982; Cairns, 1982; Flannagan and Rosenberg, 1982; Fullner, 1971; Greeson et al., 1977; Hall, 1982; Harrold, 1978; Hester and Dendy, 1962; Hellawell, 1978; Jacobi, 1971; Mason et al., 1973; McConville, 1975; McDaniel, 1974; Merritt and Cummins, 1984; Ohio EPA, 1987; Rosenberg and Resh, 1982; USEPA, 1973; Wefring and Teed, 1980.

2. Basket Sampler

A. Habitats and Substrates Sampled: All types of habitats in rivers, streams, lakes and reservoirs; may be used in areas where other methods are not feasible; not efficient for sampling in wetlands.

B. Effectiveness of the Device: Colonization depends on type of artificial substrate used in the basket (rocks, 3M Conservation Webbing, etc.); selective of certain types of fauna; three replicates considered adequate.

C. Advantages: Excellent for water quality monitoring; uniform substrate type at each station for better comparison and high level of precision; gives quantitatively comparable data; samples contain negligible amounts of debris; does not require additional weight for stability; samples a known area at a known depth for a known exposure time.

D. Limitations: Require trip for installation and another for collection; biased for insects; samplers and floats often difficult to anchor; may be navigation hazard; susceptible to vandalism; records only biotic community present during exposure period; no measure of past conditions; size and texture of limestone substrates may vary from study to study; up to eight weeks wait for results.

Selected Literature: Anderson and Mason, 1968; APHA, 1989; Benfield et al., 1974; Bergensen and Galat, 1975; Bull, 1968; Cairns, 1982; Flannagan and Rosenberg, 1982; Hall, 1982; Hanson, 1965; Hellawell, 1978; Leopold, 1970; Lium, 1974; Mason et al., 1967, 1973; Merritt and Cummins, 1984; Newlon and Rabe, 1977; Rabeni and Gibbs, 1978; Rabeni et al., 1985; Rosenberg and Resh, 1982; USEPA, 1973; Voshell and Simmons, 1977; Zillich, 1967.

5.9.5 Significance and Use of Artificial Substrate Samplers

5.9.5.1 Multiple-plate and basket samplers (Figure 9A-F) are usually colonized by a wide variety of invertebrates which have some means of mobility (active or passive) that are borne in the current. The organisms that colonize the artificial substrates are primarily aquatic insects, aquatic oligochaetes, crustaceans, cnidarians, turbellarians, bryozoans, and mollusks. The colonization of these organisms should be relatively equal in similar habitats and reflect the capacity of the water to support aquatic life. Although these samplers may exclude certain mollusks or worms, they collect a sufficient diversity of benthic fauna to be useful in assessing water quality.

5.9.5.2 Recovery techniques are critical for insuring collection of all organisms retained on the sampler.

5.9.5.3 Uniform substrate type reduces the effects of substrate differences.

5.9.5.4 Optimum time for substrate colonization is 6 weeks for most water in the United States.

5.9.5.5 Quantitatively comparable data can be obtained in environments from which it is virtually impossible to obtain samples with conventional devices.

5.9.6 Description of Multiple-Plate Samplers

5.9.6.1 Multiple-plate samplers consist of standardized, reproducible artificial substrate surfaces for colonization by aquatic organisms. Their uniform shape and texture compared to natural substrates greatly simplifies the problem of sampling. The sampler is constructed from readily available materials.

5.9.6.2 The modified multiple-plate sampler (Fig. 9A,B) is constructed of 0.125 in (0.3 cm) tempered hardboard or ceramic material with 3 in (7.6 cm) round or square plates and 1 in (2.5 cm) round spacers that have 5/8 in holes drilled in the center (Fullner, 1971). The plates are separated by spacers on a 0.25 in (0.63 cm) diameter eyebolt, held in place by a nut at the top and bottom. A total of 14 large plates and 24 spacers are used in each sampler. The top nine plates are each separated by a single spacer, plates 9 and 10 are separated by two spacers, plates 11 and 12 are separated by three spacers, and plates 13 and 14 are separated by four spacers. The hardboard sampler is about 5.5 in (14 cm) long, 3 in (7.6 cm) diameter, exposes approximately 1,160 cm^2 (.116 m^2) of surface area for the attachment of organisms, and weighs about 1 lb (0.45 kg). The ceramic sampler is 6.5 in. long and weighs 2.2 lbs (1 kg). The ceramic plates can be chemically cleaned, oven dried and reused indefinitely as they are stable and unaffected by long-term immersion in water. The sampler will not warp with time; therefore, the spacings between plates do not change, assuring replicate and efficient sampling. Each sampler is supplied with a 6 m (20′) long nylon suspension rope. The total weight is 1 Kg (2.2 lbs.). Sturdy

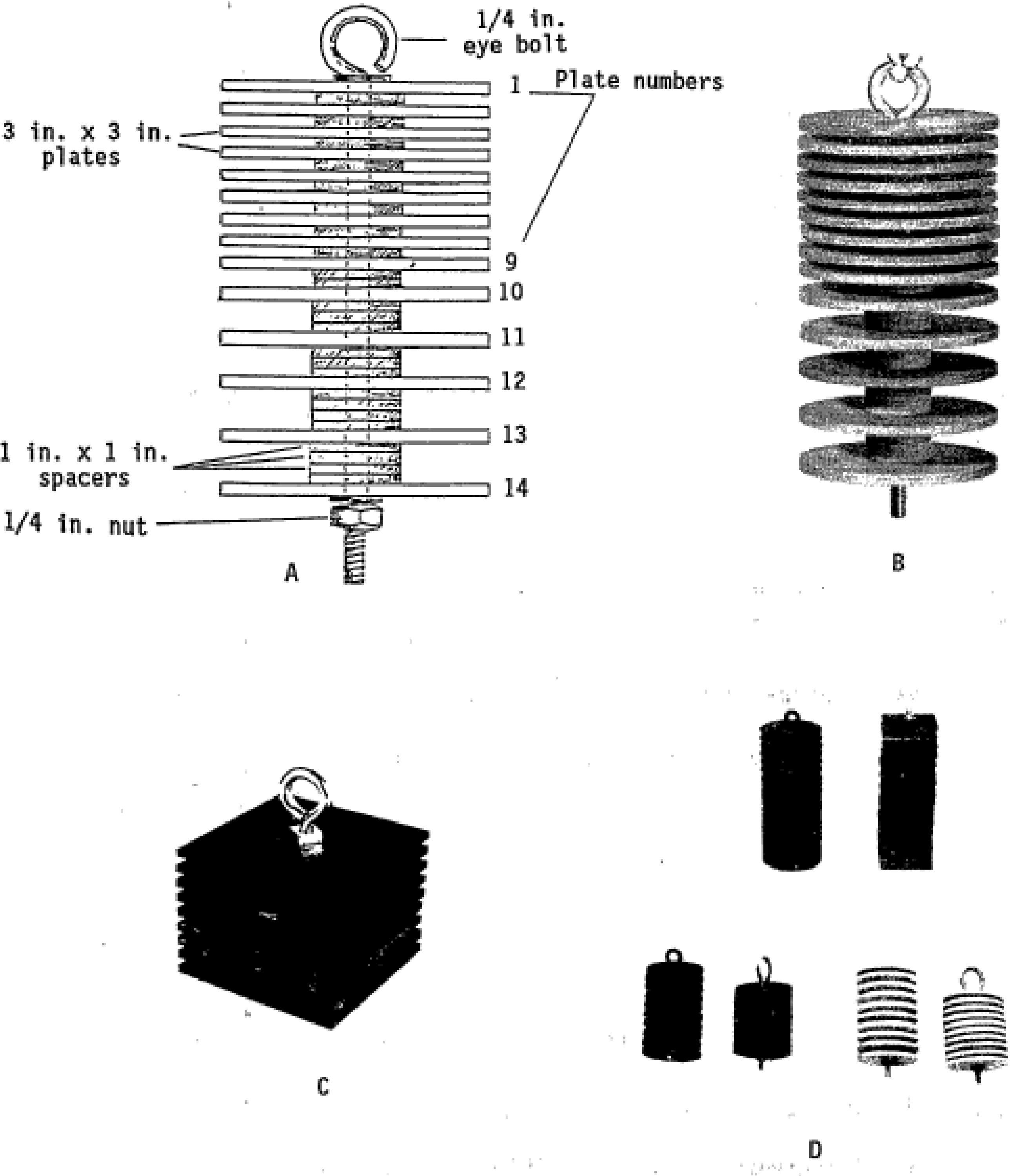
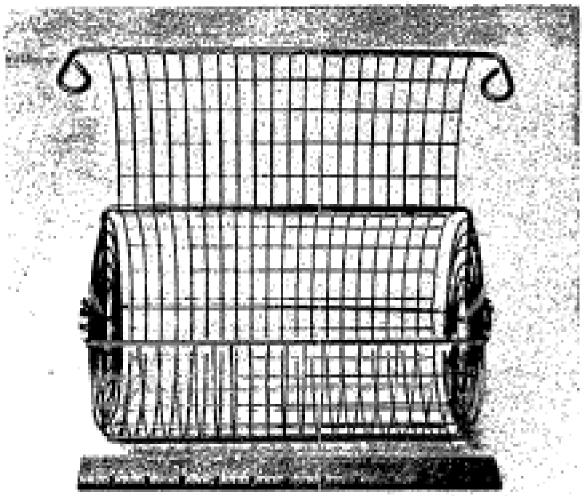


Figure 9. Artificial Substrate Samplers: (A) Schematic drawing of multiplate Sampler; (B) Typical round multiplate type; (C) Original Hester-Dendy multiplate, square design; (D) Jumbo and standard hardboard and porcelain multiplate designs



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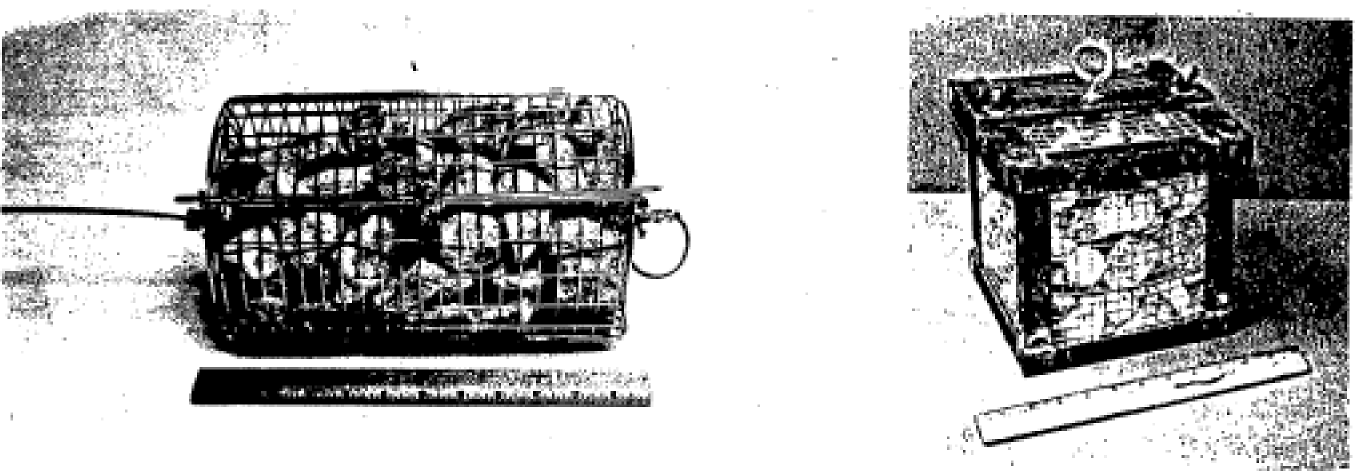

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Figure 9. Artificial Substrate Samplers: (E) Barbecue basket; (F) Basket samplers, cylindrical and square types

wire stakes for holding the sampler above the riverbed are recommended accessories.

5.9.6.3 When the samplers are suspended from the eyebolt, whether in strong currents or not, a 5 lb weight, such as a brick, is attached by .6 m wire to a 1/4 in turnbuckle. The turnbuckle is screwed tightly onto the shank of the multiplate eyebolt. The weight serves to stabilize the sampler and to lessen undue disturbance to the organisms. Upon retrieval, the weight is gently cut free before the sampler is bagged. Care should be taken not to reuse samplers exposed to oils and chemicals that may inhibit colonization during the next sampling period. Due to its cylindrical configuration, the sampler fits a wide mouth container for shipping and storage purposes. The sampler is inexpensive, compact, and light weight which are valuable attributes in water quality surveys.

5.9.7 Description of a Basket Sampler

5.9.7.1 The typical type of basket sampler (Fig. 9E) used is the one described by Mason <u>et al</u>. (1967). It is a cylindrical "barbecue" basket 11 in (28 cm) long and 7 in (17.8 cm) in diameter and is filled with approximately 17 lbs (7.7 kg) of natural rocks that vary from 1 to 3 in (2.5 to 7.6 cm) in diameter. A hinged door on the side allows access to the contents. An estimated 3.2 square ft (0.3 sq. m) of surface area is provided for colonization by macroinvertebrates. A 1/8 inch wire cable is passed through the long axis of the basket; one end is fastened with a cable clamp, and the other end is attached to a 5 gallon metal container filled with polyurethane foam used as a float. A 3/8 inch steel rod that is threaded at each end is passed through the long axis of the float and fastened at each end by nuts. Three inch long 1-1/8 by 1/8 inch strap iron secured on the rods by nuts serves as swivels at each end. The wire cable used to suspend the basket is attached to the swivels by holes drilled for that purpose. The float can be attached to a stationary structure or the basket can be anchored to the bottom in shallow water. The rugged construction of this particular basket sampler is heavy enough to resist movement by most water currents. In using the basket as a method of collecting macroinvertebrates, special consideration should be given to the types of substrates placed within the basket. Substrates tested have varied from limestone, tin cans, concrete cones, #200 3M Conservation Webbing (3M Corporation, St. Paul, MN), and porcelain spheres. Since each type of substrate will result in a different species diversity, the type of substrate used should be determined by the study objectives, weighing the advantages and disadvantages of each substrate type. For most investigations, a basket filled with 30 5-8 cm diameter rocks or rock-like material is recommended.

5.9.8 Precautions

5.9.8.1 Physical factors such as stream velocity and installation depth may variably affect degree of colonization.

5.9.8.2 The sampling method is selective for drifting organisms and for those which preferentially attach to hard surfaces.

5.9.8.3 Recovery techniques are critical for insuring collection of all organisms retained on the sampler.

5.9.8.4 Samplers are vulnerable to vandalism and often lost.

5.9.8.5 Caution should be exercised in reuse of samplers that may be subjected to contamination by toxicants, oils, etc.

5.9.8.6 The sampler provides no measure of the biota and the condition of the natural substrate at a station or of the effect of pollution on that substrate.

5.9.8.7 Sampler and floats must be anchored or fixed in place. This is sometimes difficult, and they may present a navigation hazard.

5.9.8.8 The sampler only records the community that develops during the sampling period, thus reducing the value of the collected fauna as indicators of prior conditions.

5.9.9 General Operating Procedures

5.9.9.1 Artificial substrate samplers are usually positioned in the euphotic zone of good light penetration (one to three feet, or .3-.9 m) for maximum abundance and diversity of macroinvertebrates (Mason, et al. 1973). Optimum time for substrate colonization is six weeks for most types of water in the United States. For uniformity of depth, suspend sampler from floats on 1/8 in. or 3.2 mm steel cable. If water fluctuation is not expected during sampling period, the samplers may be suspended from stationary objects. If vandalism is a problem, use subsurface floats or place sampler on supports placed on the bottom. Regardless of installation technique, use uniform procedures (e.g., same exposure period, sunlight, current velocity and habitat type). At shallow water stations (less than 1.2 m deep), install samplers so that the exposure occurs midway in the water column at low flow. If the samplers are installed in July when the water depth is about four feet and the August average low flow is two feet, the correct installation depth in July is one foot above the bottom. The sampler will receive sunlight at optimum depth (one foot) and will not be exposed to air anytime during the sampling period. Care should be exercised not to allow the samplers to touch bottom which may permit siltation, thereby, increasing the sampling error. In shallow streams with sheet rock bottoms, artificial substrate samplers are secured to 3/8 in. (.95 cm) steel rods that are driven into the substrate or secured to rods that are mounted on low, flat rectangular blocks (Hilsenhoff, 1969). These must, however, be securely anchored to the rock bottom to avoid loss during floods.

5.9.9.2 Artificial substrate samplers can be attached to floats, cement structures, a weight, or a rod driven into the stream-bed or lake-bed.

At least two or three samplers should be installed at each collecting site. Leave the samplers in place for at least 6 weeks to allow for organism colonization. The exposure time should be consistent among sites during the study. If study time limitations reduce this period, the data must be evaluated with caution, and in no case should data be compared from samplers exposed for different time periods.

5.9.9.3 The samplers may be installed in pools or riffles/runs suspended below the water surface. Make the collections as representative of the reach as possible by insuring that the samplers are not to close to the bank. In streams up to a few meters in width, install the devices about midstream. In larger streams install the devices at about one-quarter of the total width from the nearest bank.

5.9.9.4 To minimize losses of animals when retrieving multiplate and basket samplers, approach from downstream, lift the sampler quickly and place the entire sampler in a polyethylene jug or bag containing 10% formalin or 70-80% ethanol. Once the sampler is touched it must be removed from the water at once or many of the animals will leave the sampler. If the sampler must be disturbed during the recovery process so that it cannot be lifted straight up out of the water, a net should be used to enclose the sampler before it is disturbed.

5.9.9.5 The organisms can be removed in the field by disassembling the sampler in a tub or bucket partially filled with water and scrubbing the rocks or plates with a soft-bristle brush to remove clinging organisms. Pour the contents of the bucket through a No. 30 or 60 sieve and wash the contents of the sieve into a jar and preserve with 10% formalin or 70-80% ethanol. If the organisms are not removed in the field, place the sampler and the detached portion of sample into a wide-mouth container or sturdy plastic bag containing preservative for transporting to the laboratory. Label the sample with the location, habitat, date, and time of collection. The exposed multiplate sampler can be taken to the laboratory where the plates are removed from the bolt and cleaned with a soft-bristled brush. The basket samplers are usually disassembled in the field; however, they can be taken to the laboratory and disassembled if placed in preservative in a water-tight container.

5.9.9.6 Cleaned samplers can be reused unless there is reason to believe that contamination by toxicants (e.g., chemicals or oils) has occurred. These substances may be toxic to the macroinvertebrates or may inhibit colonization. Do not reuse hardboard, porcelain plates, or any other substrate that have been exposed to preservatives. Clean the multiple-plates before reassembly and use.

5.10 Coring Devices

5.10.1 Included in this category are single and multiple-head coring devices, tubular inverting devices, and open-ended stovepipe devices.

5.10.2 Selecting Coring Devices

5.10.2.1 Table 6 summarizes criteria for selecting coring devices

TABLE 6. SUMMARY CRITERIA OF CORING DEVICES

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1. KB Core Sampler

   A. Habitats and Substrates Sampled: Freshwater rivers,lakes, estuaries; soft sediments only, 40% silty clay.

   B. Effectiveness of the Device: Permits analysis of stratification in quantitative and qualitative samples; uses 5.08 cm (2 inch) pipe core tube; used in shallow to medium shallow water up to 30.5 m (100 feet) or deeper.

   C. Advantages: Samples a variety of substrates up to harder types; sampling tube can be modified for various diameters up to 100 cm^2 substrate surface; least disturbance to water/bottom interface; standard and heavy models available; wide variety of core tubes. liner tubes, core catchers, and nosepieces.

   D. Limitations: Gravity operated; samples limited surface area; standard KB core sampler head, without core tube weights approximately 8 kg (18 pounds), but additional weight can be added to sampler; requires boat and powered winch.

2. Ballchek Single and Multiple Tube Core Sampler

   A. Habitats and Substrates Sampled: Same as KB Core Sampler.

   B. Effectiveness of the Device: Samples deep burrowing organisms in soft sediment, particularly effective for sampling oligochaetes; uses 5.08 cm (2 inch) or 7.62 cm (3 inch) pipe core tube; used in shallow or deep waters, 3 m to 183 m (10-600 feet); multiple core sampler weight approximately 38 kg (84 pounds); check valves work automatically, prevent loss of sample.

   C. Advantages: Good penetration in soft sediments; small volume of sample allows for greater number of replicates to be analyzed in a short period of time; single or multiple (four) core tube sampler available; three inch pipe for larger cores and/or deep water lakes and oceans available; wide variety of core tubes, liner tubes, core catchers, and nosepieces.

   D. Limitations: Heavy device, approximately 38 kg, requires boat and winch; gravity operated; does not retain sand unless bronze core retainers are used which require additional weight to insure penetration.

3. Phelger Core Sampler

TABLE 6. SUMMARY CRITERIA OF CORING DEVICES (Continued)

A. Habitats and Substrates Sampled: Same as above core samplers.

B. Effectiveness of the Device: Similar to KB core sampler.

C. Advantages: Similar to KB core sampler.

D. Limitations: Gravity operated or can be messenger operated with a suspension-release device; styles and weights vary among manufacturers, some use interchangeable weights, between 7-35 kg, others use fixed weights up to 41 kg; length core taken varies with substrate texture.

4. Box Core Sampler

A. Habitats and Substrates Sampled: Same as above core samplers, also oceans.

B. Effectiveness of the Device: Same as above core samplers; samples a surface area of 100 cm^2 and a sediment depth of 20 cm.

C. Advantages: Same as above core samplers.

D. Limitations: Same as above core samplers; also deployed from ships or other platforms; diver collected cores are preferred.

5. Hand-Operated Core Samplers

A. Habitats and Substrates Sampled: Same as above core samplers.

B. Effectiveness of the Device: Sampled by hand or by diver.

C. Advantages: Can be used in shallow water. In deep water can be used with a diver, usually a trained biologist, who can collect and recognize substrate and bottom changes to stratify sampling; can be used with extension handles of 5, 10, or 15 feet; used with pipe fitting for driving from a pontoon boat, dock, or bridge.

D. Limitations: Limited area sampled.

Selected Literature: APHA, 1989; Brinkhurst, 1967, 1974; Burton, 1974; Coler and Haynes, 1966; Edmondson and Winberg, 1971; Flannagan, 1970; Gale, 1977; Hamilton et al., 1972; Holme, 1964; Holme and McIntyre, 1971; Miller and Bingham, 1987; Poole, 1974; Schwoerbel, 1970.

5.10.2.2 Coring devices can be used at various depths in any substrate that is sufficiently compacted so that an undisturbed sample is retained; however, they are best suited for sampling the relatively homogenous soft sediments, such as clay, silt, or sand of the deeper portions of lakes, reservoirs, and oceans. Because of the small area sampled, data from coring devices are likely to provide very imprecise estimates of the standing crop of macrobenthos.

5.10.2.3 KB type, Ballchek, and Phleger corers (Fig. 10A,B,C) are examples of devices used in shallow and deep water; they depend on gravity to drive them into the sediment. The cores are designed so that they retain the sample as it is withdrawn from the sediment and returned to the surface. Hand corers (Fig. 10D) designed for manual operation are used in shallow water. Sections of the core can be extruded and preserved separately or the entire core can be retained in the tube and processed in the field or laboratory. Intact cores can also be preserved by freezing and processed later.

5.10.2.4 Additional replication with corers is feasible because of the small amount of material per sample that must be handled in the laboratory. Multiple-head corers have been used in an attempt to reduce the field sampling effort that must be expended to collect large series of core samples (Flannagan, 1970).

5.10.2.5 The Dendy inverting sampler (Welch, 1948) is a highly efficient coring-type device used for sampling at depths to 2 or 3 meters in nonvegetated substrates ranging from soft muds through coarse sand. Because of the small surface area sampled, data obtained by this sampler suffer from the same lack of precision (Kajak, 1963) as the coring devices described above. Since the per-sample processing time is reduced, as with the corers, large series of replicates can be collected. The Dendy sampler is highly recommended for use in habitats for which it is suitable.

5.10.2.6 Stovepipe-type devices include the Wilding sampler (Wilding, 1940; APHA, 1989) and any tubular material such as 60-to-75 cm sections of standard 17-cm-diameter stovepipe (Kajak, 1963) or 75-cm sections of 30-cm-diameter aluminum irrigation pipe fitted with handles. In use, the irrigation pipe or commercial stovepipe is manually forced into the substrate, after which the contained vegetation and coarse substrate materials are removed by hand. The remaining materials are repeatedly stirred into suspension, removed with a long-handled dipper, and poured through a wooden-framed floating sieve. Because of the laborious and repetitive process of stirring, dipping, and sieving large volumes of material, the collection of a sample often requires 20 to 30 minutes.

5.10.2.7 The use of stovepipe samplers is limited to standing or slowly moving waters having a maximum depth of less than 60 cm. Since problems relating to depth of sediment penetration, changes in cross-sectional area with depth of penetration, and escapement of organisms are circumvented by stovepipe samplers, they are recommended for quantitative sampling in all shallow-water benthic habitats. They probably

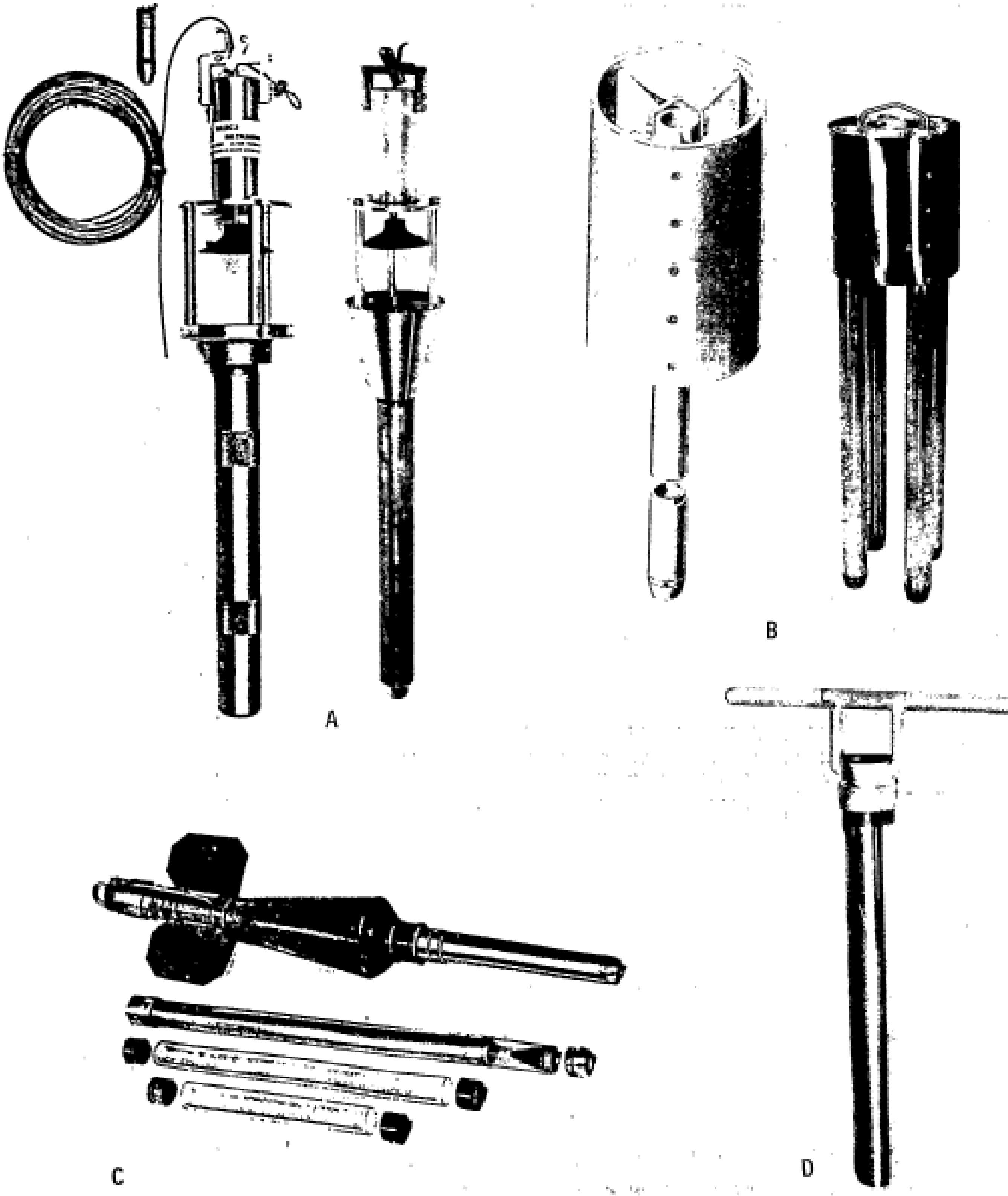

Figure 10. Core Samplers: (A) KB corer, standard and heavy duty; (B) Ballchek corer, single and multiple types; (C) Phleger corer; (D) Hand-operated corer

represent the only quantitative device suitable for sampling shallow-water habitats containing stands of rooted vascular plants and they will collect organisms inhabiting the vegetative substrates as well as those living in sediments.

5.10.2.8 In marine waters benthic macrofauna are generally collected using various box cores deployed from ships or other platforms, or diver collected cores. A box coring device consisting of a rectangular corer having a cutting arm which can seal the sample prior to retraction from the bottom should be used. In order to sample a sufficient number of individuals and species, and to integrate the patchy distribution of fauna, each sample should have a surface area of no less than 100 cm^2 and a sediment depth of at least 20 cm. In sediments having deep, burrowing fauna, a box corer capable of sampling deeper sediment may be needed. In sandier sediments, it may be necessary to substitute a grab sampler for the box corer in order to achieve adequate sediment penetration. Sufficient replicates (usually 3 to 10) should be taken to produce an asymptotic cumulative species curve. Visual inspection of each sample is necessary to insure an undisturbed and adequate amount of sample is collected.

5.11 Frames

5.11.1 For estimating the populations of attached marine organisms on a rocky shore, 0.1 m^2 or 1 m^2 square-shaped metal frames can be used for delineating percent coverage of the colonial forms. At least ten frames should be counted for characterizing the distribution statistically. Samples of the algae and macroinvertebrates should be removed from a measured area for species identification and weighed for biomass determination. It is important to note the attitude of the sampling frame relative to the horizontal and vertical axis in order to relate the data with the zonation patterns. A vertical plane is apt to have a dramatically different species array compared to a horizontal plane even with both being at the same level with the intertidal zone.

5.11.2 Attaching a 35 mm SLR camera to a sampling frame so that the focal distance is fixed is an excellent method for documenting the population present at each sampling site. Species enumeration and percent cover can be estimated from the developed photographs. This method is especially useful for documenting temporal changes at a particular sampling site.

5.11.3 For sampling the infauna of beaches, a 0.1 m^2 square metal frame with a 15 cm lip is useful. The frame can be deliberately thrown near a fixed position (see Section 4.4.3, Systematic Sampling). Stovepipe or large coffee can work very well in most sandy, sandy-mud beaches but have limited use in cobble beaches. All of the substrate is removed and screened in fine-meshed screens. The animals retained are washed or picked from the screens and preserved for later identification and enumeration.

5.11.4 Edged frames (.1 m^2) or corers can be utilized for systematically sampling the substrates around fixed positions on the flats. At least five replicate samples should be collected at each site for statistically delineating the distribution patterns of the infauna populations. The substrate is then washed through fine meshed screens. The invertebrates can be washed or picked from the screens and preserved. Flats represent areas of quiet, low velocity waters with the settling of suspended materials. Flats near pollution sources are good sites to observe the impact of all settled materials, non-toxic and toxic. Some flats are so poorly drained as to require snowshoes or similar devices for walking out to the sampling area. In such areas, it may be easier to sample at high tide from a boat using a conventional benthic grab.

5.12 Rapid Bioassessment Protocols (RBPs) for Macroinvertebrates (see Plafkin *et al*., 1989 and Section 7, Data Evaluation.)

5.12.1 The methods describe three different protocols (I,II, and III) for use in wadable streams and rivers to determine water quality. The RBPs are considered qualitative and semi-quantitative sampling techniques for assessing the health of benthic macroinvertebrate communities. The protocols consist of three basic components--water quality and physical characteristics, habitat assessment, and biosurvey. The biological assessment involves integrated data analyses of both functional and structural components of the macroinvertebrate communities through the use of metrics. The protocols describe guidelines for a rapid means of detecting water quality and aquatic life impairments and assessing their relative severity. The RBPs are not intended to replace traditional biomonitoring methods but provide an option which may be cost effective. These RBPs work very well as a surveillance tool to prioritize sites for more intensive evaluations (quantitative biological surveys) but are not always comparable to the results obtained with more traditional methods such as artificial substrate samplers or drift nets. The same metrics (RBPs) may be used with these more traditional methods of collection and give qualitative or quantitative results.

5.12.1.1 Protocol I provides for basic qualitative information for a subjective judgment of macroinvertebrate abundance and presence. The method consists of habitat assessment and the collection of macroinvertebrates from all possible habitats. The specimens are identified to orders and counted in the field. The data are used to make a subjective assessment of stream water quality or impairment.

5.12.1.2 Protocol II provides a reasonably reproducible assessment of biological impact and consists of habitat assessment and collecting macroinvertebrates from all available habitats. The specimens are identified to families, and the list of families in a 100-organisms subsample is used in the evaluation. The study is based on established guidelines in scoring parameters, and the stream site would be classified as to water quality or degree of impact and possible cause.

5.12.1.3 The objectives of Protocol III are to assess the biological impact and to establish the basis for trend monitoring of pollution effects over a period of time. The method consists also of specific guidelines for evaluating the habitat assessment parameters and collecting macroinvertebrates from all available habitats. The protocol is similar to Protocol II except that the specimens are identified to the lowest possible taxonomic level (genus, species). The data are categorized into parameters based on taxa richness, biotic index, percent composition, and functional group designations. The classification of stream sites is dependent on established guidelines.

5.13 Ohio EPA Invertebrate Community Index method (ICI) (see Ohio EPA, 1987, 1989)

5.13.1 The ICI semi-quantitative method uses 10 metrics to determine if wadable streams or rivers are polluted using benthic macroinvertebrates. Nine of the 10 metrics are based on multiple plate artificial substrate samples, and one is based on dip net sampling (Ohio EPA, 1987 and 1989). Also, see Section 7, Data Evaluation.

5.14 Standard Qualitative Collection Method (see Lenat, 1988; Eagleson, _et al_., 1990, and NC DEM, 1990 and Section 7, Data Evaluation)

5.14.1 The method emphasizes multiple-habitat sampling, field-picking of samples, and the use of both coarse- and fine-mesh samplers. This standard qualitative method consists of collecting macroinvertebrates in shallow streams, usually less than 1.5 m deep using two kick net samples, three dip net samples (sweeps), one leaf-pack sample, three aufwuchs samples, one sand sample, and visual search collections. The data resulting from this method, especially taxa richness, can be used to assign water quality ratings. The method is applicable for most between-site and/or between-date comparisons. Also, a secondary abbreviated qualitative method (EPT survey) can be used to quickly determine between-site differences in water quality. The number of collections is decreased from 10 samples in the standard quality collections to only four samples: one kick, one sweep, one leaf-pack and visual searches in the abbreviated method.

5.15 Miscellaneous Qualitative Devices

5.15.1 The investigator has an unlimited choice of gear for collecting qualitative samples. Any of the quantitative devices discussed previously, plus hand-held screens, dip nets, sweep nets, kick nets, rakes, tongs, post-hole diggers, bare hands, and forceps can be used for collecting benthic macroinvertebrates from freshwater, estuarine, and marine environments. For deep-water collecting, some of the conventional grabs described earlier and dredges are normally required. In water less than 2 meters deep, a variety of gear may be used for sampling the sediments including long-handled dip nets and post-hole diggers. Collections from vascular plants and filamentous algae may be made with a dip net, common garden rake, potato fork, or oyster tongs. Collections from floating debris and rocks may be made by hand, using forceps to catch the smaller organisms. In shallow streams, short sections of common window screen may be fastened between two poles and held in place at right angles to the water flow to collect organisms dislodged from upstream materials that have been agitated.

5.15.2 Dip, hand, sweep, kick nets and screens are rapid devices for collecting macroinvertebrates in wadable streams and rivers or at low tide in the inter-tidal zone of tidal sites. Two approaches are generally used, one in which the investigator sweeps the dip or hand net through aquatic habitats (Slack, et al., 1976; Armitage, et al., 1981) and one in which the kick net or hand held screen is held stationary against the streambed, facing upstream, and the investigator physically disturbs the stream bottom just upstream from the net or screen. The investigator vigorously kicks with the feet four or five times into the streambed to disturb the habitat in an upstream direction (Hynes, 1961; Morgan and Egglishaw, 1965; Frost, et al., 1971; Armitage, et al., 1974; Armitage, 1978; Hornig and Pollard, 1978; Pollard, 1981; and Plafkin, et al., 1989). The kicks disturb the substrate, dislodging the macroinvertebrates and some detritus, and cause the benthos to be swept by the current into the net. The debris and organisms in the kick net are then washed down into a sieve bucket and larger leaves and debris are removed.

5.15.3 Dredges are devices that are usually pulled by hand or power boat across or through the bottom sediment of a lake or stream to sample the benthos and prevent loss of active macroinvertebrates. The forward motion of the dredge carries macroinvertebrates into the net.

5.15.3.1 Elliott and Drake (1981a,b) compared four light-weight dredges for sampling in rivers. They indicated that the dredges are not suitable for quantitative sampling. Also, considerable variation existed in their effectiveness as qualitative samplers for estimating the total number of taxa per sample.

5.15.3.2 Dredges should be emptied after collection into a shallow tray, bucket, or sieving device if the sample is sorted on-site. The sample can be placed directly in labeled wide-mouth containers with preservative and transported back to the lab for processing.

5.16 Suction Samplers

5.16.1 Suction samplers have been used widely in sampling macroinvertebrates in fresh, estuarine, and marine waters (Brett, 1964; Larsen, 1974; Gale and Thompson, 1975). They can be placed directly on the sampling station and can be operated by hand in shallow water or by a scuba diver in deep water (see 5.18).

5.17 Photography

5.17.1 The use of photography is mainly limited to environments that have suitably clear water and are inhabited by sessile animals and rooted plants. Many estuarine habitats, such as those containing corals, sponges, and attached algal forms, fall in this category and can be photographed before, during and after the introduction of stress. The technique has been used with success in south Florida to evaluate changes brought about by the introduction of heated effluents.

5.17.2 The technique for horizontal underwater photos using scuba gear involves placing a photographically identifiable 1.0 m^2 area frame or marker in the habitat to be photographed and an additional nearby marker on which the camera is placed each time a photograph is taken. By this means, identical areas can be photographed repeatedly over a period of time to evaluate on-site changes in sessile forms at both affected and control stations. Vertical, overhead photos may be taken under suitable conditions.

5.17.3 Photographs are also useful in documenting a habitat or alterations in a station over time (e.g., increase in canopy cover, changes in channelization of a stream, and effects of flooding, etc.).

5.18 **Scuba**

5.18.1 This equipment can be used in freshwater sampling of mollusks in large riverine systems or with diver collected cores.

5.18.2 The reader is referred to Simmons (1977), Sommers (1972), U.S. Department of the Navy, U.S. Navy diving manual (latest edition), and Gale and Thompson (1974) for much additional information on this subject. All USEPA diving operations should be conducted in accordance with standards set forth in the U.S. EPA Occupational Health and Safety Manual-1440, 1986, entitled Chapter 10, EPA Diving Safety Policy. Therefore, if the need for diving capability exists, approval must be obtained through an USEPA regional laboratory diving officer. Scuba gear can be used to improve aquatic sampling; in particular sampling of mussels, other benthos, and fish. Isom, et al., (1979) reported utilizing scuba in rediscovery of snails, which were thought to be extinct. Various investigators had sampled the same areas previously on numerous occasions.

5.18.3 Gale (1977) notes the numerous applications of scuba to sampling benthos including placement and retrieval of artificial substrate; use of suction samplers (Larsen, 1974; Gale and Thompson, 1975); sampling with a quadrate frame; and, perhaps most importantly, identifying and delineating substrate types for purpose of determining sampling effort (stratified sampling) and choice of samplers.

5.18.4 If pelecypods (freshwater mussels) are to be sampled with brails in areas which historically contained them and/or it is desired to sample quantitatively, scuba can be used effectively in taking quadrates. In large rivers, which have mussel beds with homogenous substrate, it is desirable to take at least 10 square meter quadrates (10,000 square cm each). In small rivers where the mussels' niche may be between rocks and it is generally difficult to place a square meter frame, then a 0.5 square meter frame (2500 square cm) should be utilized with no less than 3 square meters, or twelve 0.5 square meter samples taken. Samples should be taken randomly in all cases, which in the latter instance, will result in collection of good representative diversity (see Section 2, Quality Assurance and Quality Control).

5.18.5 Scuba diving is safe if conducted by rigid safety standards, some of which are mandatory for scientific/educational diving (See Federal Register, July 22, 1977; 42, 41: pp. 37650-37673). Conformance with these and subsequent standards is costly but essential for safe conduct of scuba sampling. See references listed above for more in depth discussion of safety, the buddy system, etc. The need for observance of safety rules cannot be overemphasized.

5.19 Brails

5.19.1 This device is primarily limited to sampling of bivalve mussels in large (non-wadable) rivers.

5.19.2 The use of brails for commercial harvest of mussels has been the common practice since before 1900; however, this practice and scuba have been used by investigators to study mussel populations on a limited basis.

5.19.3 The reader is referred to Coker (1919), Van der Schalie (1941), Scruggs (1960), Lopinot (1967), Isom (1969), Bates (1970), Starrett (1971), and Buchanan (1980) for more information on collecting mussels, brails, and brailing. Coker (1919) describes how to make a brail.

5.19.5 Once the site to be sampled has been identified, reference should be made to historical literature for determination of species that may be encountered.

5.19.6 Quantitative sampling is accomplished with a crowfoot brail to determine the rate of catch per drag from a given area. All equipment can be made or rented from and fished by a commercial fisherman. Each brail sample consists of dragging a measured distance of 100 m, then sorting and counting the catch. The area sampled is calculated in square yards by multiplying the length of brail by 100 m. Catch success is expressed in terms of the average catch of mussels per square per drag. Brail sampling is randomized within fishing area and by time periods during two complete harvest seasons (March through August).

5.19.7 Brailing is also an effective qualitative sampling device, especially in large, deep rivers. Where possible, the services of a commercial mussel fisherman should be utilized. The experienced mussel fisherman is adept at using brails and only extensive experience would make an investigator's results equivalent to the general mussel fisherman. Maximum legal brail length is 16 feet (approximately 5 m) in some states; diameter of wire used for hooks is also controlled. These points can be worked out with the state permitting agency.

5.19.8 A minimum of six 100 m long hauls (drags) should be accomplished where a single brail is used. Most commercial fisherman use two brails simultaneously; thus, only three hauls would be required. Record the time for each haul; however, take about 20 minutes to make each haul since a very slow speed is best for catching mussels. If the hauls are made too fast, the catch will be small. If a significant mussel population is found, then qualitative or quantitative scuba (see 3.18, Scuba) samples should be taken. A minimum of 10 m^2 samples should be taken by scuba at each station. All specimens should be identified to species, growth cessation rings counted, and measured for determination of population age structure.

5.19.9 Mussel fishing with brails is highly dependent on experience of the user; however, they are very efficient in the hands of experienced users as attested to by almost 100 years of continuous use.

5.19.10 Availability of brailing equipment may be a deterrent to its use; however, if the method is adopted more widely by the scientific community, suppliers may develop to meet the need.

5.20 Other Mussel Collecting Methods

5.20.1 Mussels found in small or medium sized streams and rivers that can be waded are often found most numerous on bars where the pools break off into shoals. Sometimes, there are constrictions in streams at these points where weed beds can be found. Sample into the lower end of pools, around the weed beds, and in riffles/runs and fast-flowing water. A long-handled rake modified with a rectangular collection basket of one-quarter inch wire mesh, dredge dip net, or using the hands are the best method for sampling mussels from these habitats (Starrett, 1971). It is advisable to wear gloves and place a net below the area being sampled to catch small mussels that might otherwise not be collected.

5.20.2 Other collection techniques and procedures can be found in the 1941 Annual Report of the American Malacological Union. Information on collecting snails can be found in the same publication.

5.20.3 If rare or endangered species are collected, they should be returned to their habitat since it is illegal to take such species.

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